MONITORING NEKTON IN SALT MARSHES

(Revision #1)

A Protocol for the National Park Service's Long-Term Monitoring Program, Northeast Coastal and Barrier Network

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The Protocol Narrative

This protocol is an adaptation of the protocol developed by Raposa and Roman (2001a) for use in the Long-term Coastal Monitoring Program at Cape Cod National Seashore. The original protocol can be found that the National Park Service Inventory and Monitoring website: http://www.nature.nps.gov/im/monitor/protocoldb.cfm. Extensive portions of text have been borrowed from Raposa and Roman (2001a) and are presented in this document.

Protocol Background

National Park Service (NPS) managers need accurate information about the resources in their care. They need to know how and why natural systems change over time, and what amount of change is normal, in order to make sound management decisions. Therefore, the National Park Service has begun natural resource monitoring throughout the National Park System to gather this information as part of the Natural Resource Challenge program. A key component of this effort, known as Park Vital Signs Monitoring, is the organization of approximately 270 park units into 32 monitoring networks to conduct long-term monitoring for key indicators of change, or "vital signs." Vital signs are measurable, early warning signals that indicate changes that could impair the long-term health of natural systems. Early detection of potential problems allows park managers to take steps to restore ecological health of park resources before serious damage can happen.

This protocol describes the methodology used to sample nekton (fish and crustaceans) in shallow (<1m) estuarine habitats within and adjacent to salt marshes such as salt marsh pools and adjacent shoreline areas as part of the NPS Park Vital Signs Monitoring Program. Estuaries and the wetlands that fringe them are critical habitat for wildlife and perform many valuable services. Since estuaries are the link between land and sea many of the practices on land (agriculture, industry, and urban and residential development) can directly impact the quality of estuarine resources and ecosystems. Threats to estuarine ecosystems include eutrophication, watershed development, wetland loss, overfishing, and other human-induced problems. Long-term monitoring of nekton is especially valuable for addressing questions related to long-term/large-scale ecosystem changes and processes. Monitoring estuarine natural resources, such as nekton, is needed to document the effects of anthropogenic impacts, to follow trends in natural changes (*e.g.*, sea level rise), and to provide baseline datasets that can be used for natural resource damage assessment in the case of catastrophic events such as oil spills. Developing and initiating long-term nekton monitoring programs will help track natural and human-induced changes in estuarine nekton over time and advance our understanding of the interactions between nekton and the dynamic estuarine environment. Additionally, long-term data are useful for differentiating natural and human induced variability and for formulating testable hypotheses regarding the ecology of estuarine species (Wolfe *et al.* 1987).

Nekton, defined here as an assemblage of fishes and crustaceans (such as shrimp and crabs), is an abundant estuarine fauna with unique responses to environmental change

that make them desirable for inclusion in a coastal monitoring program. There are many factors that make nekton a potentially useful and informative monitoring variable in estuaries (Neckles and Dionne 2000; Neckles *et al*. 2002; Raposa *et al*. 2003). Fig. 1 identifies some of the linkages between human-induced and natural environmental stressors (*e.g.,* altered hydrology, nutrient enrichment, storms, and sea level rise), associated changes in estuarine habitat structure, and responses of the nekton community. Estuarine nekton is an integral link among primary producers, consumers, and top predators and is likely to respond to either top-down or bottom-up estuarine perturbations. For example, nutrient enrichment (a bottom–up perturbation) could affect nekton by altering submersed vegetative habitats (Valiela *et al.* 1992; Harlin 1995). Conversely, removal of predatory fishes through overfishing (top-down) could induce responses in the forage or prey nekton guild (Carpenter and Kitchell 1985). Nekton also represents a significant portion of the diets of many piscivorous birds, economically valuable fishes, and, when in estuaries, marine mammals (Friedland *et al.* 1988; Sekiguchi 1995; Smith 1997). Long-term monitoring will also document the introduction or expansion of invasive species (*e.g.*, Japanese shore crab, *Hemigrapsus sanguineus*), interactions among invasive and native species and their subsequent impact on nekton community dynamics, and changes in species ranges. Development of the Index of Biotic Integrity (Karr 1981) and the Estuarine Index of Biotic Integrity (Deegan *et al.* 1997) attests to the value of monitoring nekton to document ecosystem level responses to anthropogenic stress. The foundation of these indices lies in the notion that fishes and crustaceans incorporate and reflect multiple ecosystem processes, and therefore indicate overall ecosystem integrity.

Shallow water salt marsh habitats are especially important to include in a nekton monitoring program. Sampling in salt marsh habitats is emphasized due to the susceptibility of each habitat to anthropogenic stress and to the abundant and rich nekton assemblages that each habitat supports. Salt marshes are an important habitat for nekton, including juveniles of economically valuable species in some regions (Deegan 1993; Able *et al.* 1996; Kneib 1997; Minello 1999; Roman *et al.* 2000). Salt marshes provide food and refuge for estuarine species and there is evidence that they enhance the productivity of estuarine nekton assemblages (Boesch and Turner 1984). The position of nekton in the upper levels of marsh food webs as well as their dependence on a wide variety of food resources and habitats serve to integrate salt marsh processes and ecosystem elements (Kwak and Zedler 1997). Nekton responds to ecosystem changes resulting from anthropogenic impacts. For example, fish abundance, species richness, and growth rates of the mummichog (*Fundulus heteroclitus*) increased in response to enhanced nitrogen loading (LaBrecque *et al.* 1996; Tober *et al.* 1996). Several studies have also indicated that nekton responds rapidly (*e.g.,* within days to months) to the manipulation of salt marsh hydrology (Rey *et al.* 1990; Taylor *et al.* 1998; Able *et al.* 2000; Roman *et al.* 2002).

Salt marshes have also been heavily impacted by human activities, including extensive mosquito grid ditching (Bourn and Cottam 1950; Daiber 1986) and restriction of tidal flow by roads, causeways, and culverts (*e.g.*, Roman *et al.* 1984 and 1995; Rosza 1995; Burdick *et al.* 1997; Dionne *et al.* 1999). Today, extensive efforts are underway to restore natural tidal regimes to these degraded marshes by removing tide-restricting

structures, excavating new habitats such as creeks and pools, and planting marsh grasses. Monitoring nekton is one way of documenting the response of natural communities and marsh functions to restoration efforts.

This protocol has been developed for shallow subtidal habitats (<1m) that retain water throughout the tidal cycle. And more specifically, this protocol is intended for sampling shallow habitats within salt marshes (*e.g.*, creeks, pools) and shallow subtidal habitats immediately adjacent to salt marshes. The methodology in this protocol is not appropriate for sampling nekton within estuarine intertidal flats, deep eelgrass beds, or gravel/rocky substrates. Information gained from monitoring nekton should augment concurrent monitoring of other estuarine resources and processes. For example, monitoring only nekton would not comprehensively describe the effects of environmental change (such as sea level rise or restoration) but monitoring nekton along with vegetation, bird use, hydrology, and other variables would provide a more complete view of ecosystem responses to environmental change and enable an evaluation of linkages among habitat characteristics and trophic levels.

Protocol Objectives

Specific objectives and monitoring questions addressed by the Nekton Protocol have been developed in association with those for the salt marsh vegetation and salt marsh elevation: :

Objective 1: To understand long term changes in salt marsh vegetation and nekton communities.

- **Question 1:** *Are salt marsh vegetation patterns (species composition and abundance changing over time (e.g., decades)?*
  - **Vital Sign 1:** Salt Marsh Vegetation Community Structure
- **Question 2:** *Is nekton community structure (species composition, abundance, and size structure) changing over time (e.g., decades)?*
  - **Vital Sign 1:** Salt Marsh Nekton Community Structure

Objective 2: To understand responses of salt marsh vegetation and nekton communities to environmental change.

- **Question 1:** *How do salt marsh communities change in response to perturbations (e.g., invasive species, oil spills, storms) in the environment?*
  - **Vital Sign 1:** Salt Marsh Vegetation Community Structure
  - **Vital Sign 2:** Salt Marsh Nekton Community Structure

Objective 3: To understand how salt marsh elevations respond to local sea-level rise.

- **Question 1:** *Are salt marsh surface elevation trajectories changing over time (e.g., decades), and if so, what factors are contributing to observed elevation changes (e.g., surface versus subsurface processes, changes in organic matter accumulation)?*
  - **Vital Sign 1:** Salt Marsh Sediment Elevation

- **Question 2:** *Are salt marsh surface elevation trajectories keeping pace with the local rate of sea-level rise?*
  - **Vital Sign 1:** Salt Marsh Sediment Elevation

Species composition and abundance of nekton responds to environmental changes (*e.g.*, sea level rise, nutrient loading, invasive species colonization). Monitoring nekton over time will help evaluate natural and human-induced changes in estuarine nekton in the long-term and will advance our understanding of the interactions between nekton and the dynamic estuarine environment.

Protocol History

The original protocol (Raposa and Roman 2001a) was developed at Cape Cod National Seashore, an NPS prototype monitoring park. Development of this protocol was based on quantitative data (Raposa 2000; Raposa and Roman 2001a; Raposa and Roman 2001b; Raposa *et al*. 2003) that were collected from sampling programs in five southern New England estuaries (Fig. 2, Table 1). From these data, guidelines for the temporal and spatial frequency of sampling, appropriate replicate sample size, and appropriate statistical analyses were developed. The result was the development of the original protocol entitled *Monitoring Nekton in Shallow Estuarine Habitats* authored by Raposa and Roman (2001a).

As part of a pilot program to implement the nekton protocol within the National Park Service, this protocol has been used at 7 National Parks (as of 2004) within the Northeast Coastal and Barrier Network (NCBN) and the Northeast Temperate Network (NETN) (Fig. 3). In the summer of 2003, the protocol was tested at Colonial National Historical Site (COLO), Fire Island National Seashore (FIIS), and Gateway National Recreation Area (GATE). In 2004, the protocol was tested at Cape Cod National Seashore (CACO), Sagamore Hill National Historic Site (SAHI), Boston Harbor Islands National Park Area (BOHA), and Saugus Iron Works National Historic Site (SAIR). Additional pilot studies are scheduled to begin in 2005 at Assateague Island National Seashore (ASIS), George Washington's Birthplace National Monument (GEWA), and possibly Acadia National Park (ACAD).

Data collected from the nekton protocol will help address issues and concerns not only at the estuary, park, and Network levels but also at the Regional level. The General Conceptual Model (Fig. 4) for the Coastal and Barrier Network identifies major external activities or processes that influence the natural system (Agents of Change), the associated problems or products of human activities or natural events that alter the quality or integrity of the ecosystem (Stressors), and the measurable changes in ecosystem structure, function, or processes (Ecosystem Processes). Since nekton responds, often quickly, to environmental change, a program that monitors estuarine nekton will be able to detect changes in species composition and abundance, or shifts in trophic relationships, providing an early warning system to larger ecosystem threats or alterations.

Protocol Summary

This protocol describes the methods used to sample nekton (free-swimming fish and crustaceans) in shallow water (<1m) habitats such as salt marsh pools, tidal creeks, and

shallow shoreline areas adjacent to salt marshes and is a revision of the Nekton Monitoring Protocol developed by Raposa and Roman (2001a). The following recommendations for sampling procedures follow those put forth by Raposa and Roman (2001a); however, there are also updates to the protocol listed herein, most notably the inclusion of the ditch net as a sampling gear. Study sites are selected using a stratified random approach. Sampling stations are randomly located within pools and along ditches, creeks, or shoreline areas. Nekton is sampled exclusively with throw traps in shallow water salt marsh habitats (creeks, pools) and ditch nets in narrow mosquito ditches. There should be two daytime sampling efforts per year; one in early summer (June-July) and another in late summer-early fall (August-October), unless there are species or processes unique to other seasons that are of interest. A minimum of 15 replicate throw trap samples and 10 replicate ditch net samples (depending on the availability of habitat) should be collected from each marsh during each sampling event. Nekton composition, and the density and length of individuals from each species are recorded at each sampling station. Environmental parameters are collected concurrent with nekton sampling include temperature, salinity, water depth, dissolved oxygen, and vegetation cover.

This protocol is presented as a minimum for nekton monitoring. If additional time, personnel, or funds are available, supplementary sampling can be initiated; for example, additional sampling in spring, concurrent sampling on the marsh surface with a bottomless lift-net, or measurements of nekton biomass. There are also some limitations associated with the design. For example, sacrificing more sampling dates in favor of a large sample size during two sampling periods increases the possibility of missing short-term pulses of migrating species or newly hatching young-of-the-year. It would also not be possible to estimate growth rates by tracking modal lengths of cohorts over time. If growth rates (or production) were of interest, then a research or monitoring program with more sample dates would be appropriate.

Sampling Design

The sampling design of the Nekton Protocol has been developed after extensive research and sampling in the field. The rationale for the sample design is discussed in detail in Raposa and Roman (2001a) and is briefly presented in this section. The following questions have helped shape the development of this protocol, and the sampling design and methods for the nekton protocol are best described in terms of these questions.

What is the population of interest?

The populations of interest are those of estuarine fishes and crustaceans (nekton) that are either residents or transients of the selected salt marsh monitoring sites and their adjacent shallow water shoreline areas in the coastal parks of the NCBN and NETN. These fishes and crustaceans are relatively small, with most individuals less than 100mm in total length. They include permanent resident fishes of salt marsh ecosystems (*e.g.,* killifish, sticklebacks, minnows) and crustaceans (*e.g.,* shrimp, crabs) or transients that use estuarine salt marshes as nursery grounds during the summer months (*e.g.,* herring, flounder, eels).

What is measured?

The measurements of importance in the Nekton Protocol are the total density of nekton and the density and size of individuals for each species. Every individual captured is identified to species and counted. Additionally, up to 15 randomly selected individuals of each species are measured (mm) for either total length (fish and shrimp) or carapace width (crabs). When a large number of throw trap samples are collected (*e.g,*. >25), mean lengths obtained by measuring only 5 individuals per trap sample did not differ from mean lengths when 30 individuals were measured (Raposa and Roman (2001a). This was true for three different types of species: a decapod (*Palaemonetes pugio*), a ubiquitous-high density fish (*Fundulus heteroclitus*), and a patchy-high density fish (*Menidia menidia*). Although accurate length estimates can be obtained by measuring as few as 5 individuals per throw trap sample, we suggest a more conservative approach by randomly measuring at least 15 individuals of each species, particularly if distinct cohorts (*e.g.,* young-of-the-year and adults) are present or if analyses of trends in life history stages are desired. By measuring nekton lengths, information can be gained on habitat use by different life history stages. For example, by measuring mummichog (*Fundulus heteroclitus*) sizes from Cape Cod National Seashore's Hatches Harbor salt marsh throw trap samples, Raposa and Roman (2001a) demonstrated changes in the size distributions (from seasonal sampling data) of this species throughout the year, emphasizing the influx of young-of-the-year in summer. These data will provide estimates of nekton density, species richness, community composition, and length frequency distribution. Additionally, other statistics such as species richness can be calculated from these data.

The protocol strongly urges that additional environmental data are also recorded. Measuring associated environmental variables when collecting nekton will help define the sampling environment during monitoring. Certain variables may change with anthropogenic impacts over time; for example, lower dissolved oxygen levels with increased macroalgae from nutrient enrichment, increased salinity with tidal restoration, or conversely, decreased salinity with impoundment. By concurrently sampling basic measures, researchers can better define causal mechanisms for observed temporal changes in nekton (Raposa and Roman 2001a). At each nekton sampling station, the following environmental data are recorded: water temperature (ºC); water salinity (ppt); dissolved oxygen (mgl^{-1}); water depth (cm); ditch depth (cm) (for ditch net stations only); and the estimated percent vegetative cover (using cover class categories).

What is the appropriate sampling unit?

The sampling unit is an enclosure trap (either throw trap or ditch net) that traps nekton species within a known surface area, therefore allowing for abundance and density calculations. The throw trap measures 1m square by 0.5m high and thus encloses a known area ($1m^2$). The ditch net measures 1m long by 1m deep, but since it is flexible it can sample any size ditch up to 1m wide and 1m deep. The surface area that the ditch net samples is calculated from measuring the distance between the corners of the net. Both of these gears provide quantitative estimates of nekton abundance (number of fish per m^2).

How many samples should be taken?

Densities of estuarine nekton are highly variable, especially over spatial scales (Raposa and Roman 2001a; Raposa *et al.* 2003). One way to address this variability and improve the ability to detect biological differences (*e.g.,* species richness, density) among treatments is to increase sample size. Determining the appropriate sample size depends on a number of factors, such as the desired level of precision, the type of statistical comparisons (if any) that are to be made, and the desired difference among treatments one wishes to detect (Krebs 1989; Sokal and Rohlf 1981). Sample size also varies among different nekton species and depends on different attributes of the nekton community that are under consideration (*e.g.,* density, richness).

A power analysis was conducted for the original protocol to determine the appropriate sample size for sampling estuarine nekton with the $1m^2$ throw trap (Raposa and Roman 2001a; Raposa *et al.* 2003). The objective of this power analysis was to determine the minimum number of sample replicates that are necessary to detect changes in species composition between nekton communities of salt marshes. Power is a function of the differences between two populations, the sample size, the alpha level of the test (the probability of detecting a difference between two datasets when no difference exists, *i.e*., Type I Error), and the variability of the measured response. The results of the power analyses are shown in Fig. 5. In this figure the horizontal axis indicates the similarity or "sameness" of two different nekton communities (using Euclidean distance as a similarity index) with those communities that are similar at the left portion of axis and those that are different on the right portion of axis. If only 5 replicates are taken at each site, there is low power to detect differences, even for those cases where the differences are great. Increasing the sample size to 15 dramatically increases the power to differentiate two nekton data sets, even between data sets that are quite similar. With a power above 0.9, there is a >90% chance of detecting a difference between data sets when a difference actually exists. With a low power there is an increased probability of not detecting a difference when the data sets are actually different (*i.e*., Type II Error). If subtle differences in nekton density are of interest (*e.g*., comparing nekton density in the same marsh from one year to the next), or if one is interested in detecting differences within individual species between sampling years, then it may be appropriate to have a large number of replicates. If detecting only dramatic changes were the objective (*e.g*., comparing pristine Marsh A with highly impacted Marsh B), then perhaps a smaller number of replicates would suffice (Raposa and Roman 2001a; Raposa *et al*. 2003). In order to maximize power for the multiple analyses that will be conducted as part of this protocol while still maintaining a reasonable level of field sampling effort for a crew of 4 in one day, 15 throw trap samples should be taken. For marshes with fewer than 15 pools, all pools should be sampled.

We recommend at least 10 ditch net samples should be collected from each site during each sample period. As of this writing an extensive analyses of capture efficiencies and replicate sample sizes for the ditch net has not yet been performed. The replicate size of 10 is a our best estimate based on the power analysis of the throw trap data and the logistics of sampling one marsh with a crew of at least 4 people in one day. However, we present this method as an ancillary method to sample ditches where throw traps cannot be used.

How should sampling units be positioned?

Throw traps should be used to sample salt marsh pools, larger tidal creeks, and shoreline areas of salt marshes. Salt marsh pools should be at least $2m^2$ in surface area to sample, as it is difficult to precisely throw the trap (so the trap lands entirely within the pool) in pools that are smaller. Throw trap stations are randomly assigned to pools within the marsh and the specific station location within a pool is randomly located along the pool's perimeter. If there are fewer than 15 pools on the marsh, then all pools are sampled. If there are more than 15 pools, then 15 pools are randomly selected as station locations. Typically, only one station per pool is desired, however, on larger pools two or three stations may be sampled as long as the stations are further than 30m apart. The exact station location on a pool is randomly located along the perimeter of each pool. Locations of throw trap stations along shoreline areas are randomly located along the length of the shoreline. Adjacent stations should be at least 30m apart. If closer placement of stations is necessary to achieve adequate replicate size, then adjacent stations must be sampled at least 30min apart.

Ditch nets are used to sample grid ditches and smaller tidal creeks of salt marshes. Ditches should be at least 15cm wide (to allow free passage of nekton through the net prior to triggering) and have between 10cm and 1m depth of water when triggered. Ditch nets are randomly located along the length of the ditch or tidal creek. Ditch nets should be at least 30m apart. Since ditch nets must be sampled within a critical window of the tidal period, sampling adjacent stations 30min apart is not an option if stations are closer than 30m.

When will the samples be taken?

Spatial variability in nekton abundance is much higher than temporal variability in freshwater systems due to habitat heterogeneity (Peterson and Rabeni 1995). These authors found that collecting a larger number of samples on fewer dates would optimize sampling efforts, as opposed to taking a smaller number of samples spread out over multiple dates. To our knowledge, a similar detailed analysis of spatio-temporal variability does not exist for estuarine nekton. However, an analysis using nekton densities in tidal creeks from three southern New England salt marshes suggests that variability patterns may be similar for estuarine nekton (Raposa and Roman 2001a). Temporal variability in density among sampling dates was on average 21 times smaller than spatial variability (*i.e.*, variability among samples taken on the same sampling date). Because of this, we adopt the sampling strategy suggested by Peterson and Rabeni (1995) and suggest that a larger number of samples be collected on fewer dates to address spatial variability and improve sampling precision.

Monitoring estuarine nekton is dependent on the tidal cycle of the marsh. Nekton sampling should occur at the same relative tide stage. The timing of sampling is more critical for ditch net samples than for throw trap samples. A thorough reconnaissance of the study site and its specific tidal regime should be well documented prior to sampling.

Sampling in subtidal salt marsh habitats (*e.g*., creeks and pools) with a throw trap should occur only after the marsh surface is drained of tidal water (low or ebbing tide or prior to flood tide). If the marsh surface is flooded during sampling, densities of species that utilize the marsh surface will be underestimated in subtidal habitats.

Sampling in narrow tidal creeks or ditches with a ditch net should occur when water has drained off the surface of the marsh, but when there is still enough water in the ditches and creeks to sample (more than 10cm in depth). The timing of sampling for ditch nets is very critical, since the nets need to be set at least 30min prior to sampling, to allow disturbance from setting up the nets to dissipate. If the nets are set too late into an ebbing tide, the ditches will be drained before the nets are sampled.

Some studies have demonstrated differences in estuarine nekton composition and abundance between day and night periods (Rountree and Able 1993, Heck *et al.* 1989). Using throw traps at Hatches Harbor, Raposa and Roman (2001a) documented significantly higher densities of green crabs (*Carcinus maenas*) at night. However, densities of all other species were not different between day and night at Hatches Harbor, and this protocol recommends that samples only be collected during the day. This approach should provide accurate representations of the densities of most species in the study sites, keeping in mind that some species, due to their diurnal rhythms (particularly decapods), may be underrepresented during the day. The logistics of daytime sampling are more accommodating for field personnel and day sampling facilitates comparisons with a larger number of datasets. However, night sampling could be initiated in the future to augment regular daytime sampling if time and resources allow, or if a particular question can only be addressed by night sampling.

The highest nekton density and richness occurs during warm weather temperatures in temperate estuarine habitats (Pearcy and Richards 1962; Recksiek and McCleave 1973; Adams 1976; Cain and Dean 1976; Hoff and Ibara 1977; Orth and Heck 1980; Pihl and Rosenberg 1982; Pihl Baden and Pihl 1984; Ayvazian *et al*. 1992; Rountree and Able 1992; Able *et al*. 1996; Lazzari *et al*. 1999; Raposa and Roman 2001a; Raposa and Roman 2001b). In some cases the exact timing of nekton peaks depends on latitude and/or habitat type. For example, nekton abundance in eelgrass beds peaked in June in Chesapeake Bay (Heck and Orth 1980, Orth and Heck 1980), but peaked in late summer and fall in Nauset Marsh (Heck *et al.* 1989). In Cape Cod and other southern New England salt marshes, abundance peaked in landward habitats (marsh pools, upstream tidal river) later in the year than in seaward habitats (marsh creeks, downstream tidal river) (Raposa and Roman 2001a).

Despite the variability in the timing of abundance and richness peaks, both density and richness are generally highest between June and October in temperate estuaries and monitoring efforts should be concentrated during this period to maximize information gained per sampling effort. Therefore, this protocol recommends sampling nekton twice per year, once in early summer (after June 15) and in late summer-early fall (August to early October). Sampling prior to June 15, in the Northeast is not recommended because water temperatures are still cold and few nekton will be collected. The time frames for

sampling nekton will vary due to differences in climates in the Network's region, for example nekton in Maine will be sampled between June 15 and September 15, whereas sampling in Virginia is recommended from June 1 through October 15. Each sampling effort for each park should be concluded within 7 to 10 days.

Should sampling units be permanent or temporary?
Sampling station locations within salt marsh habitats remain permanent for the sampling year, but from year to year should be re-randomized. Re-randomization between sampling years is preferred because station markers frequently are disturbed during the winter months or can be lost entirely if sampling occurs over a 3-5 year intervals, and re-locating stations is time consuming.

It is possible that the same pool and ditches will be sampled during different sampling years (especially if there are fewer than 15 pools on the marsh). However, since the station location on the pool or ditch is re-randomized between years, the same sampling location within the pool or along the ditch (*i.e.*, microhabitat) is different between years.

What sites are sampled?
Study sites will be selected using a stratified random sampling design, if more than two sites are available within the park. For example, if the there is an extensive stretch of salt marsh (such as at FIIS or ASIS) the entire salt marsh system will be stratified and sampling locations will be randomly selected within each stratum. An example of stratification that might be used would be distance from an inlet. To be selected for monitoring, a study site must meet the following criteria: it must be representative of the larger salt marsh system in which it occurs, there must be adequate nekton habitat (marsh pools, creeks, shoreline) area to allow for a minimum of 15 replicate sampling stations, and the site must be accessible. In addition, it is useful to co-locate sites where the proposed Salt Marsh Vegetation and Salt Marsh Sediment Elevation Table monitoring protocols will be implemented. These additional data will provide insights on processes influencing the entire salt marsh ecosystem and thus the nekton community.

For many NCBC and NETN parks there are fewer than two salt marshes within the park (*e.g.,* BOHA, GATE, GEWA, SAIR, SAHI). In these instances, there are only one or two areas to sample, and those areas will be monitored.

As of the summer of 2005, we have sampled salt marsh vegetation using this protocol at several National Park Service sites. Sites within parks were selected as follows.

Assateague Island National Seashore (ASIS): Study locations were randomly selected within strata of grazing intensity by ungulates (*i.e.*, ponies). Grazing (an important resource issue at ASIS) intensity strata were low grazing, moderate grazing, and high grazing. Areas of grazing intensity were identified by ASIS Resource Management staff, and overlaid with grid (500m by 500m grid cells) in GIS. All grid cells were numbered and three grid cells ($500m^2$) were randomly chosen from the population of available grids within each strata. Three random grid cells were chosen as it was necessary to have back-up grid cells if logistical issues (i.e. access to sites) or co-

location of other sampling efforts (*i.e.*, SETs) prevented the use of a particular randomly selected grid. Nekton locations at ASIS are an unnamed marsh (moderate intensity grazing), and Valentines Marsh (high intensity grazing). The high intensity site (North End marsh) was not sampled for nekton as there were no salt marsh pools at this site. Maps of study locations will be included after stations have been sampled.

Boston Harbor Islands National Park Area (BOHA): Thompson Island marsh was sampled in 2004 (Figs. 6 & 7). This was the only salt marsh within BOHA that had sufficient open water habitat to sample.

Colonial National Historical Site (COLO): Back River Marsh (on Jamestown Island) and Kings Creek Marsh (on the York River) were sampled in 2003 (Figs. 8 & 9). Back River Marsh was chosen as a sampling location because resource management required information on the marsh for the Jamestown Project (C. Rafkind, pers. comm). Kings Creek was chosen as a representative estuarine salt marsh for COLO. This site was specifically chosen due to access issues at other sites.

Fire Island National Seashore (FIIS): Hospital Point and Watch Hill Marshes were sampled in 2003 (Figs. 10 & 11). Sediment Elevation Tables (SETs) were already established at both sites and it was decided to co-locate nekton sampling with the SETs. The marsh area where the SETs were located was chosen using a stratified random design with distance from the inlet as the stratification (C. Roman, NPS, pers. comm.). Access to the site was also a consideration for the SET locations.

Gateway National Recreation Area (GATE): Horseshoe Cove marsh within the Sandy Hook Unit and Big Egg marsh within the Jamaica Bay Unit were sampled in 2003 (Figs. 12 & 13). Horseshoe Cove is the only marsh on Sandy Hook of sufficient size to sample the required number of nekton stations. Additionally, Sediment Elevation Tables (SETs) were already established at Horseshoe Cove and it was decided to co-locate nekton sampling with the SETs. Big Egg marsh is currently undergoing restoration (vegetation is being monitored by GATE staff, G. Frame, NPS, pers. comm.) and nekton data are being collected to aid in the evaluation of the restoration.

George Washington Birthplace National Monument (GEWA): There are only two tidal salt marshes within GEWA. These marshes are Pope's Creek (including the islands within Pope's Creek) and Dancing Marsh. Due to the small size of both marshes, the entire marsh area is the study site. Maps of study locations will be included after stations have been sampled.

Saugus Iron Works National Historic Site (SAIR): The Saugus River adjacent to the salt marsh was sampled in 2004 (Fig. 14). This is the only open water within the park.

Sagamore Hill National Historic Site (SAHI): The salt marsh adjacent to Cold Spring Harbor was sampled in 2004 (Figs.15 & 16). This is the only salt marsh within the park.

Sampling Methods

What equipment should be used for sampling?

The recommended sampling gear for monitoring nekton in salt marsh and shallow (<1m) estuarine ecosystems are enclosure traps (throw traps and ditch nets). Enclosure traps are quantitative sampling gear that have a high and consistent capture efficiency in most habitats, tend to better represent benthic nekton, and are small enough (typically 1 m^2) to permit sampling in specific microhabitats (Zedler 1990; Rozas and Minello 1997). No gear can effectively sample the entire nekton assemblage in all habitats, but the high and consistent capture efficiency is a primary advantage of throw traps over seines. Higher capture efficiencies may also lower sample variance, and thus, sample size during monitoring (Peterson and Rabeni 1995).

Throw traps and seines sample a different area of habitat per unit effort. Most throw traps sample 1m^2. However, a small 10m seine covers almost 80m^2 in a single quarter-circle haul. Because they sample a larger area, seines might be expected to collect more species than traps. However, during this protocol's development, estimates of species richness using throw traps (13.9 species) and seines (16.9 species) in tidal creeks in a Cape Cod salt marsh were not significantly different (Student's t-test; $p>0.05$; Raposa 2000; Raposa and Roman 2001a). Furthermore, the smaller creeks and pools of salt marshes can only be sampled by throw traps as seines are too big (Raposa 2000). Narrow creeks, small pools, and grid ditches are utilized by nekton and are important habitats that would go undocumented when sampling with only a seine. For these reasons, we concur with Rozas and Minello (1997) and suggest using throw traps for monitoring nekton in shallow (< 1m) estuarine habitats.

We present two gear types to sample estuarine nekton, depending on the habitat. The preferred gear is the 1m^2 throw trap which can be used to sample salt marsh pools, tidal creeks, and shoreline areas. The second gear is the ditch net which can be used to sample mosquito ditches and smaller tidal creeks (<1m wide). The throw trap is a 1m^2 box (measuring 1m^2 wide x 0.5m high) that is open the top and bottom. The sides of the trap are covered with 3mm mesh hardware cloth. Since the trap samples a known area of water (1m^2) quantitative and repeatable estimates of nekton density can be obtained. A 1m^2 throw trap is best used within sand or mud bottomed estuarine habitats. In gravel or rocky bottoms the seal between the trap bottom and the substrate is often not tight and capture efficiency decreases.

To adequately describe the nekton community within mosquito ditches the ditch net is the gear of choice. The ditch net (not described in Raposa and Roman 2001a) is an enclosure gear designed to sample narrow mosquito ditches and smaller tidal creeks up to 1m wide and 1m deep within salt marshes. Grid ditches are common features on salt marshes. These ditches, which were created for mosquito control purposes in the 1940's, vary in width from 45cm to 100cm and on some marshes, especially those in southern New England, are the only water habitat within the marsh. The 1m^2 throw trap cannot adequately sample these narrow ditches. Density can be calculated for a ditch net by measuring the area of water the net is sampling. The center body of the net lines the

sides and bottom of 1 linear meter (approximately) of ditch. There are two doors on the open ends of the net, which when pulled, rise up to close off the ends of the net, enclosing an area of water that is 1m long and as wide as the ditch.

The primary rationale for selecting the ditch net as a sampling gear for narrow tidal ditches is that no other sampling gear can sample this environment. This gear has been adopted by GPAC part of their regional standards and protocols for monitoring restoration sites in the Gulf of Maine Region (GPAC Workshop 2004). The throw trap is not a good sampling gear for the grid ditch habitat, as the trap is too large. Even smaller versions of a throw trap would not sample these areas effectively as the trap would have to land precisely in the ditch in order to enclose the nekton. Seines cannot be used as the ditches are very narrow (45cm wide) and therefore the net cannot be properly deployed. Fyke nets (bottom-anchored nets consisting of mounted netting bags) could be used to sample these habitats, however the area the fyke net samples is difficult to determine, and thus only species composition, and not density, can be measured.

Field personnel

At least two field technicians are required to physically conduct the field sampling at a maximum of 2 to 3 sites. Since monitoring nekton requires close coordination with the specific tidal regimes of sampling sites (which may only occur only two weeks of every month), it is advised that primary responsibility of the field technicians be nekton monitoring rather than using technicians assigned to other duties to "fill-in" for nekton sampling. However, it is possible to piggy-back other monitoring protocol responsibilities (such as the salt marsh vegetation protocol) with the duties of the technicians assigned to the nekton protocol, if scheduling is carefully mapped out prior to the sampling season. For example, during the initial testing phase of this protocol we used the same technicians to monitor nekton and salt marsh vegetation at all sites each summer.

Preparation prior to field sampling

Prior to the field season, all sampling gear should be checked and repaired if necessary. All electronic equipment (*e.g.,* GPS, water quality probes) should be calibrated and tested prior to sampling in the field and field personnel should be trained use all equipment. A complete reconnaissance of field sites should be made at several different tidal stages so that information on tidal cycles, flooding regime, and site geography (*i.e.* suitability of pools and ditches for sampling) can be documented and a schedule can be developed. Sampling stations should be located and marked in the field prior to the first sampling. Maps of the sampling site should be made prior to sampling. The maps should have all station locations clearly marked. If boat access is required to reach sampling sites, arrangements should be made well in advance of the first sampling.

Since the time frame for sampling nekton is dependent upon the tides (nekton should be sampled after the marsh surface has drained of water), it is imperative that all sampling events be scheduled prior to the sampling season. This is especially important if more than one marsh is being sampled, as tidal cycles may only allow appropriate sampling windows two weeks of each month. Scheduling sampling for ditch nets is much more

important in terms of timing than for throw traps. The ditch net method requires that there be water present in the ditches to sample, and since the net must be deployed at least 30min prior to sampling, the timing can be critical especially on marshes that drain very quickly.

Conducting sampling

Once the sampling schedule has been arranged, sampling is relatively easy. This protocol urges that two or more teams of people be used for sampling efficiency and safety reasons, although one team of two people can accomplish sampling at a limited number of sites. The station location on the pool is randomly located along the perimeter of the pool prior to sampling. To use the throw trap, the sampling station is quietly approached and the trap is tossed into the pool, shoreline area, or tidal creek. The trap is then pressed down into the sediment to prevent nekton from escaping from under the trap. All nekton within the trap are collected (using a dip net), identified, and 15 randomly selected individuals from each species are measured. All nekton are returned alive back into the pool, creek, or shoreline area. All data are recorded in the field on field data sheets (examples of field forms are provided Section 2). Two people are required to sample the ditch net. The ditch net is set up at least 30min prior to sampling. The net is set in the ditch, suspended by four stakes. The stakes should be pushed into the sediment so they are stable, but not so hard that they are difficult to extract. The doors of the net are pushed down into the bottom of the creek, so as not to impede the passage of nekton through the net, and the lines from the doors are laid out on the marsh surface. The dimensions between the stakes are measured (to calculate the area of water that is sampled). After 30min, each person quietly approaches the lines to the doors that have been laid out on the marsh surface. Each person then simultaneously pulls on the lines, causing the doors of the net to rise and enclose a portion of the water column. As the lines are being pulled, the net is approached, and once the doors are completely up, the stakes are grabbed and pulled from the ditch, trapping all nekton in the net. The net is then laid on the marsh surface, and all nekton within the net are identified, and 15 randomly selected individuals from each species are measured. All nekton are returned alive back into the ditch. All data are recorded in the field on field data sheets.

For both methods, voucher specimen(s) of any unknown or questionable identification should be retained, humanely sacrificed (by a quick blow to the head), placed in 70% ethanol, and transported back to the laboratory for positive identification.

Associated environmental variables (water temperature, salinity, dissolved oxygen) should be recorded after each station is sampled. Vegetation cover within the throw trap, if present, should be estimated (as percent cover) prior to dip netting fish, as dip netting may disturb the vegetation and influence cover estimates.

Data Management

Data should be entered into the Northeast Coastal and Barrier Network Monitoring Program Salt Marsh Database (Access software program) as soon as possible after collection. Any unknown specimens should be identified immediately upon return to the laboratory and the correct identification indicated on the field datasheet. Any edits,

changes, or corrections to the data should be noted on the field data sheet and include the date and person (initials) verifying the change or correction. All GPS coordinates should be entered into a GIS program (*e.g.*, ArcView) to verify the locations of sampling stations and to provide maps of sample stations for the second round of nekton sampling.

Analysis and Reporting

Data collected from the nekton monitoring should be summarized yearly by each monitoring site. Local and regional analyses should be conducted at every 5 year intervals and include all data and all parks monitored to date. All data are stored in an Access database (Northeast Coastal and Barrier Network Monitoring Program Salt Marsh Database) and reports can be generated from this database. Additional summaries and analyses may require the export of data from the Access database into other programs.

Data summaries and statistical analyses

Annual Reports

Routine data summaries to be presented in reports include species composition (species lists), average total nekton density, and total number of individuals collected (Table 2). These summaries will be easily available from the reporting form section the Access database (Northeast Coastal and Barrier Network Monitoring Program Salt Marsh Database). It may also be of interest to report average densities of fish and crustaceans separately, or densities and size distributions of individual species (Table 3). Averaged lengths of nekton measured and averages for environmental variables should also be reported (Fig. 14, Table 4). An estimate of error (standard error or standard deviation) and sample size (number of stations sampled or number of individuals measured) should be presented in all tables and graphs (if appropriate).

Trend Reports

When data from more than one site or more than one sampling year have been collected, statistical analyses will be conducted to determine if nekton density, nekton length frequency distributions, or community structure is changing over time. An Analysis of Variance is used to determine if nekton densities for a specific site are changing over time. Distribution tests, such as the Kolmogorov-Smirnov test, are used to determine if size-frequency distributions of a specific species are changing over time. Changes in community structure (species composition and abundance) can be assessed by using, for example, analyses (*e.g.*, ANOSIM) that are part of the PRIMER (Plymouth Routines In Multivariate Research, Carr 1997) software package, (http://www.primer-e.com), that use non-parametric permutation procedures to detect differences in community structure. ANOSIM is just one example of a non-parametric test, similar to multivariate analysis of variance (MANOVA) but without the generally unattainable assumptions (Clarke and Warwick 1994, Carr 1997). Non-parametric permutation testing procedures can be effectively used to evaluate dissimilarity or similarity in nekton communities between marshes or between sample years. In the typical analysis ANOSIM (Analysis of Similarities) is used to determine if there are differences in community structure either among years or between sites.

Reporting Schedule

Reports presenting monitoring information for parks that were sampled, data summaries, statistics (if applicable), and any problems or special circumstances/events that were encountered are reported on a yearly basis and submitted to the each park's Natural Resource Manager and the NCBN coordinator (Bryan Milstead, Bryan_Milstead@nps.gov). Reports should be generated in a timely fashion and be submitted no later than the spring following the monitoring season (*e.g.*, monitoring for summer 2004 should be reported by May 2005).

A trend analysis report will be generated for every 5 years of data. This is a comprehensive report that includes a Network and regional overview of the monitoring program, management plans, summaries of all data to date, statistical comparisons among years (if appropriate), any concerns or problems, and suggestions to improve or augment the existing monitoring program. The first trend report is due in 2008 and will include all data collected from 2003 to 2007, the next trend report would be due in 2013 and would include all new data from 2008 to 2012 as well as a trend analyses (*e.g.*, ANOSIM) for the entire dataset, with all subsequent reports following this same timeline. The most important component of the trend report is the analysis of the long-term monitoring data for each site and park. Trend analysis reports are submitted to each park's Superintendent, and Natural Resource Chief and the NCBN coordinator (Bryan Milstead, Bryan_Milstead@nps.gov).

Operational Requirements

Operational requirements for the implementation of the nekton protocol include a schedule for park units and sites, staff to conduct sampling and oversee aspects of the monitoring and data analyses, and funds for supplies and travel expenses.

Personnel

Personnel required for implementation of the Nekton Protocol are one supervisor and at least 2 field technicians. The supervisor oversees all aspects of the monitoring from coordination with parks, to the initial study site selection, station location, sampling schedule, equipment manufacture and repair, data collection, species identification and verification, data entry, and data analyses (if applicable). At least two field technicians are required to physically conduct the field sampling at a maximum of 2 to 3 sites.

A minimum of two people are required to sample nekton in the field, but four or six people are recommended. It is useful to have one person who is the lead person in the sampling endeavor. This person can instruct other personnel on what needs to be done prior to and during the sampling season as well as making sure that all equipment are in working order and that data are correctly recorded. It is desirable to have personnel who are familiar with estuarine fishes and their identification. However, since there are only approximately 20 or so species that will be collected, it is also possible to train personnel on the job, as most species are easy to identify. It is strongly suggested that each park initiate contacts with fisheries experts at local universities, colleges, or other agencies, in case that further expertise is required in the identification of unknown or rare species.

All personnel should be physically fit, able to spend long hours in field conditions (hot and humid weather, walking on uneven ground), and be able to carry field equipment.

Scheduling

The implementation schedule for NCBN and selected parks within the Northeast Temperate Network is presented in Table 5. During the pilot testing phase of the nekton protocol 3 to 5 park units (each with 1 to 3 study sites) were sampled twice each year (in June and August) by a crew of 4 field technicians. Additionally, these technicians also were able to collect vegetation cover data as part of the salt marsh vegetation monitoring program. One supervisor oversaw the pilot implementation phase and was responsible for obtaining research permits, maintaining contact with each Park's Natural Resource Manager, overseeing data collection, data quality control, data entry, and reporting.

The nekton protocol should be implemented every 3 years at each specific long-term monitoring site. After the testing phase in 2003 – 2005, parks are sampled every 3 years. Technicians could be shared among parks that are in the same geographic region (*e.g.*, ASIS, COLO and GEWA or FIIS, GATE and SAHI). If this is done, these technicians must be dedicated to the sampling for the monitoring protocol(s) in order to effectively monitor all sites.

A team of four technicians can sample more sites, and this is an option if more than one park within the same geographic region is monitored within the same year. The technicians could be shared among the parks thus accomplishing monitoring at several sites within one year. This may be a more cost effective method than having the technicians located at on central location and traveling to the monitoring sites which can be costly. However, this may require regional oversight of the monitoring program from year to year to ensure adequate supervision training, quality control of the data, and reporting responsibilities.

Testing of the nekton protocol started in 2003. We tested both the nekton and salt marsh vegetation protocol at the same time, and thus the field crew was responsible for collecting both nekton and vegetation data. Nekton were sampled in June and August, while vegetation was sampled July. We found this to be a very efficient, but somewhat taxing for the field crew (primarily due to extensive traveling to and from sites), method for accomplishing both nekton and vegetation monitoring at several sites within one sampling season.

The tidal regime of sampling sites is the limiting factor to the number of potential field days, and thus the amount of sampling that can occur. If sampling sites have the same tidal regime, the number of possible sites sampled may decrease, since only two weeks of every month will have tides favorable to sampling. Conversely, if the tidal regimes rarely result in the marsh surface flooding at some sampling sites, these sites will have more acceptable sample days and more sites can be sampled.

Four people can efficiently sample one site (*i.e.*, 15 throw trap nekton stations) in one day. If only 2 people are sampling, the number of sampling days required per site is

increased. Sampling using the ditch nets requires more time than sampling with the throw trap, as the nets must be set up prior to sampling. If timed appropriately, both the ditch nets (10 stations) and throw trap (15 stations) samples can be sampled by 4 people in one to two days. The benefit of having a dedicated field crew is that there will always be enough help to conduct the sampling. The downside of a dedicated field crew is that for weeks where tides are not favorable to sampling, there may be little for them to do. Additionally, if nekton is sampled only in June and August, there will be no work for the crew in July. This is why we implemented both the nekton and vegetation protocols in the same year.

Budget

The budget for implementation of the nekton protocol includes the salary for at least 2 full time seasonal (May through August or September) field technicians (GS level 4 to 7, depending on qualifications) and part time salary for one supervisor (approximately GS level 10 or higher).

Budget for supplies is minimal, especially if water quality equipment (*e.g.*, water probes, YSI, refractometers, thermometers) is already owned by the park or Network and is available for use by the field technicians. If this equipment is not available then it must be purchased. A standard YSI unit will cost approximately $1500 to $2500 depending on the model. If funds are not available for the purchase of a YSI, dissolved oxygen could be dropped as an associated environmental variable and a simple thermometer and refractometer (approximately $200) can be used to measure temperature and salinity.

Both the throw trap and ditch net are easy and relatively inexpensive to manufacture, and once built can be used year after year, if maintained. Throw traps cost approximately $150 and ditch nets cost approximately $50. However, some supplies such as netting, leadcore line, and rope may only be available through bulk purchase, so sharing costs and materials for sampling gears among parks and building several of each gear type may be the most cost-effective option. Ditch nets require more upkeep, since netting is more prone to rips than the wire mesh of the throw trap. Additional tools (*e.g.*, drills, drill bits, hammers, sewing skills, *etc*.) are also required to build the sampling gears.

Other miscellaneous supplies that are required are hip boots for field personnel (approximately $100 per pair), a few vials to store voucher specimens, fish identification guides, field notebooks (we prefer waterproof notebooks or waterproof paper for data sheets), clipboards, oak stakes or flags for marking sampling locations, and permanent markers. Having maps of sampling stations, preferably in GIS form, are a great help in setting up and locating stations in the field.

If technicians are traveling to several sites then funds must be budgeted for travel expenses and a reliable vehicle must be available for transportation. Occasionally other travel expenses such as vessel time are also required, as in the case of BOHA. As an example, vessel time to and from the islands of BOHA cost approximately $80 per hour (total vessel expense for the 2004 sampling season for BOHA was $400).

Version Control Procedures

This protocol is a revision of a protocol first developed by Raposa and Roman (2001a) for use in the Long-term Coastal Monitoring Program at Cape Cod National Seashore. The original protocol can be found that the National Park Service Inventory and Monitoring website: http://www.nature.nps.gov/im/monitor/protocoldb.cfm

This protocol was revised for the following reasons:

- To conform to NPS format guidelines
- To include the additional sampling method of the ditch net sampler

Previous Version	Revision Date	Author	Changes Made	Reason for Change	New Version #
Original Protocol	12/13/04	Mary-Jane James-Pirri mjjp@gso.uri.edu	Format Changes; Addition of ditch net SOP	Conform to NPS guidelines; Add ditch net as gear type	#1

Table 1. Sites and sampling regimes at five estuaries in southern New England. Sampling at all sites was conducted with throw traps only. Data were used from two distinct sampling programs at Galilee.

	Hatches Harbor	**Herring River**	**Nauset Marsh**	**Sachuest Point**	**Galilee**	**Galilee**
Location	Provincetown, MA	Wellfleet, MA	Eastham, MA	Middletown, RI	Narragansett, RI	Narragansett, RI
Geographic coordinates	42º06' N 70º23' W	41º 57' N 70º 04' W	41º 50' N 69º 57' W	41º28' N 71º14' W	41º22' N 71º30' W	41º22' N 71º30' W
Habitats sampled	Creeks, pools	Tidal channel	Marsh edge, eelgrass, creeks, pools	Creeks, pools	Creeks, pools	Creeks, pools
Sampling period	6/97-6/98	5/98-2/99	5/98-2/99	1997-1999 (Aug-Oct)	1997-1999 (Jun-Sep)	8/98-5/99
Sampling frequency	Biweekly	Seasonally	Seasonally	Monthly	Monthly	Seasonally
Total samples	770	240	500	300	392	160

Table 2. Average density [number m^{-2} ± SD (total count)] of nekton sampled from ditches (with ditch nets) and pools (with throw traps) at Hospital Point and Watch Hill marshes, FIIS, in 2003. Replicate sample size is given after site name.

Species	Common Name	Hospital Point (36)	Watch Hill (29)
Ditches			
Cyprinodon variegatus	Sheepshead minnow	0	6.3 ± 28.3 (28)
Fundulus heteroclitus	Mummichog	0.9 ± 2.2 (7)	17.3 ± 41.8 (106)
Fundulus majalis	Striped killifish	0	0.6 ± 2.1 (4)
Lucania parva	Rainwater killifish	0	0.2 ± 0.6 (1)
Palaemonetes pugio	Grass shrimp	0	6.1 ± 27.3 (27)
Pools			
Anguilla rostrata	American eel	0.1 ± 0.5 (2)	0.1 ± 0.3 (1)
Apeletes quadracus	4-spine stickleback	0.2 ± 0.9 (4)	0.3 ± 0.7 (3)
Crangon septemspinosa	Sevenspine bay shrimp	0.1 ± 0.2 (1)	20.7 ± 27.6 (186)
Fundulus heteroclitus	Mummichog	1.2 ± 2.5 (21)	2.6 ± 6.9 (23)
Fundulus majalis	Striped killifish	0.1 ± 0.2 (1)	0
Fundulus species	*Fundulus* species	0.1 ± 0.2 (1)	0
Goby species	*Goby* species	0	0.1 ± 0.3 (1)
Menidia menidia	Atlantic silverside	2.5 ± 7.4 (45)	0.7 ± 1.4 (6)
Palaemonetes pugio	Grass shrimp	30.1 ± 86.9 (541)	1.7 ± 5.0 (15)
Syngnathus fuscus	Northern pipefish	0	0.2 ± 0.7 (2)

Table 3. Average density [number m^{-2} ±SD (total count)] of fish and decapods sampled from ditches and pools at FIIS in 2003. Replicate sample size is given after site name.

Variable	Hospital Point (36)	Watch Hill (29)
Ditches		
Total Fish	0.9 ± 2.2 (7)	24.5 ± 67.0 (139)
Total Decapods	0	6.1 ± 27.3 (27)
Total Nekton	0.9 ± 2.2 (7)	30.5 ± 92.1 (166)
Pools		
Total Fish	4.2 ± 8.6 (45)	4.4 ± 10.9 (36)
Total Decapods	30.1 ± 87.1 (542)	22.3 ± 31.1 (201)
Total Nekton	34.2 ± 95.0 (587)	26.3 ± 39.5 (237)

Table 4. Physical characteristics (average ± SD) of nekton sampling stations (ditches and pools) at FIIS in 2003. Replicate sample size is given after site name.

Variable	Hospital Point (36)	Watch Hill (29)
Ditches		
Water Temperature	22.3 ± 3.9	23.2 ± 1.3
Salinity	14.4 ± 7.1	15.1 ± 7.6
Dissolved Oxygen	2.1 ± 2.5	4.3 ± 2.7
Pools		
Water Temperature	24.3 ± 5.8	25.3 ± 2.8
Salinity	13.8 ± 7.6	20.0 ± 7.2
Dissolved Oxygen	1.9 ± 2.0	7.8 ± 1.1

Table 5. Suggested sampling schedule for NCBN parks. * Indicates that some parks may be monitored more frequently due to special circumstances (*e.g.*, ongoing restoration).

Year/Park	2003	2004	2005	2006	2007	2008	2009	2010
ASIS			**X**			**X**		
ACAD			**X**			**X**		
BOHA		**X**			**X**			**X**
CACO		**X**			**X**			**X**
COLO	**X**		**X**			**X**		
FIIS	**X**			**X**			**X**	
GATE*	**X**	**X**		**X**			**X**	
GEWA			**X**			**X**		
SAHI		**X**		**X**			**X**	
SAIR		**X**			**X**			**X**

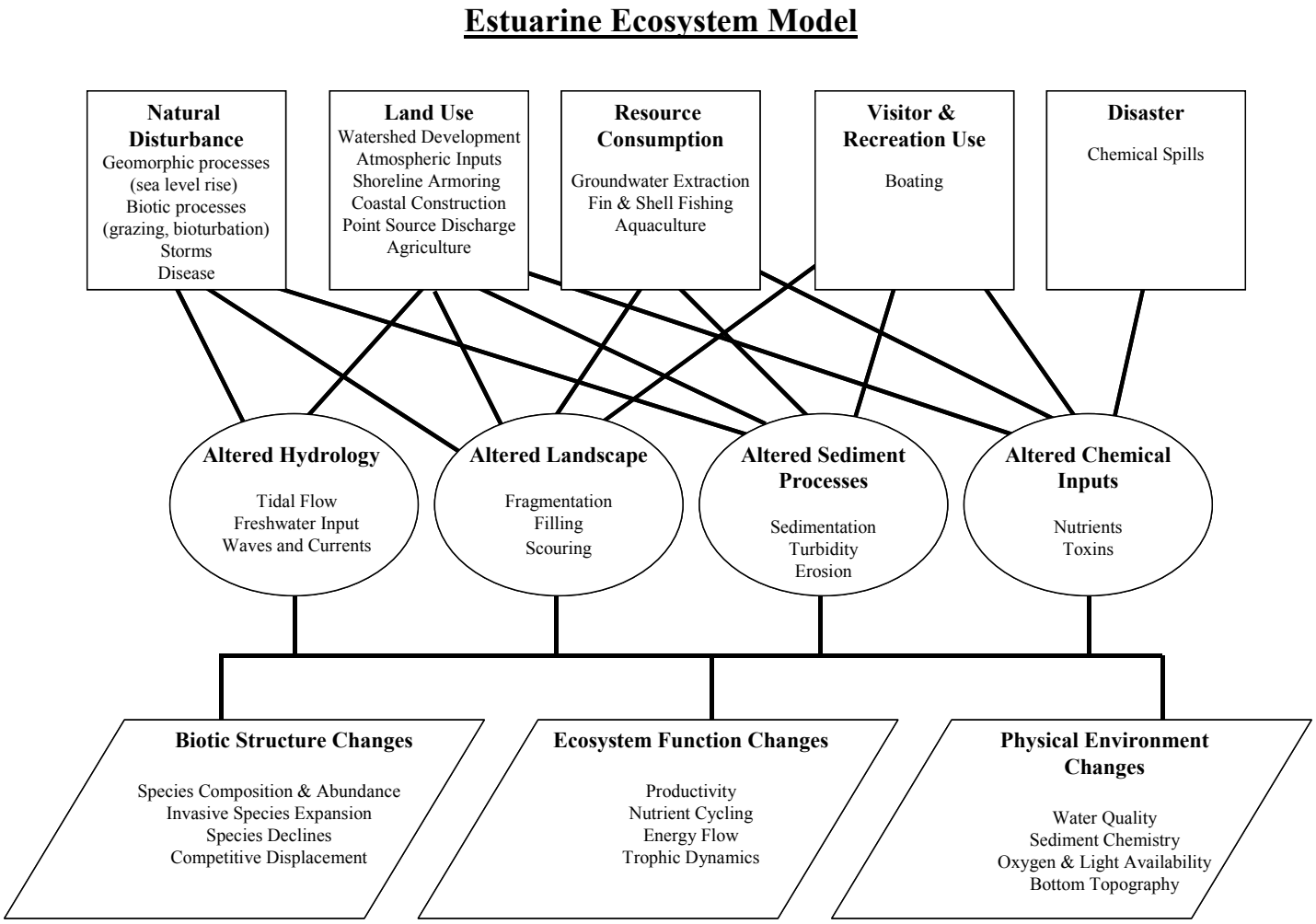


Figure 1. The Northeast Coastal and Barrier Network Estuarine Ecosystem Model.

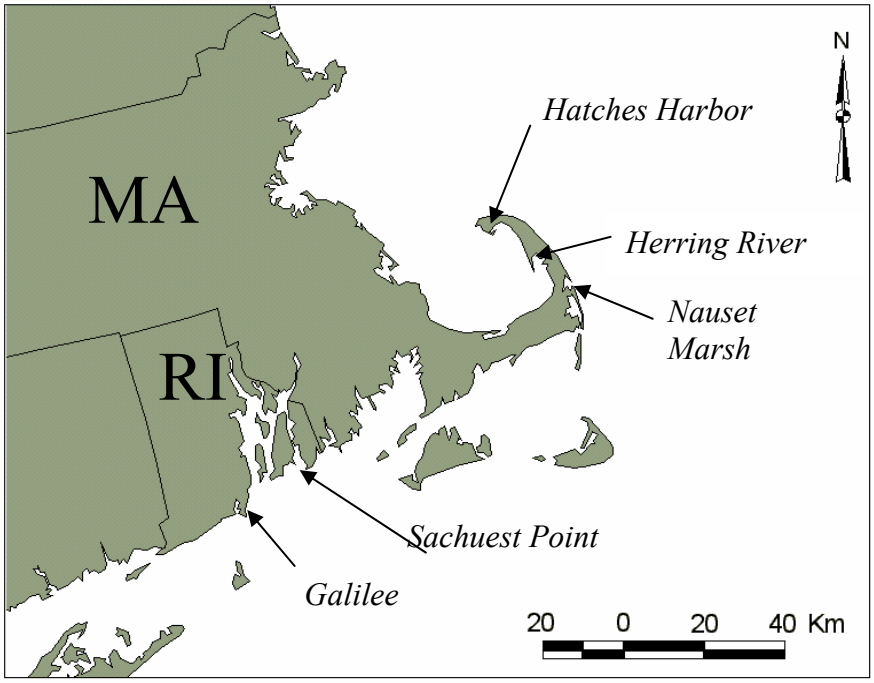


Figure 2. Sites of the five study sites in southern New England where nekton throw trap data were collected from 1997-1999 as part of the development of the protocol.

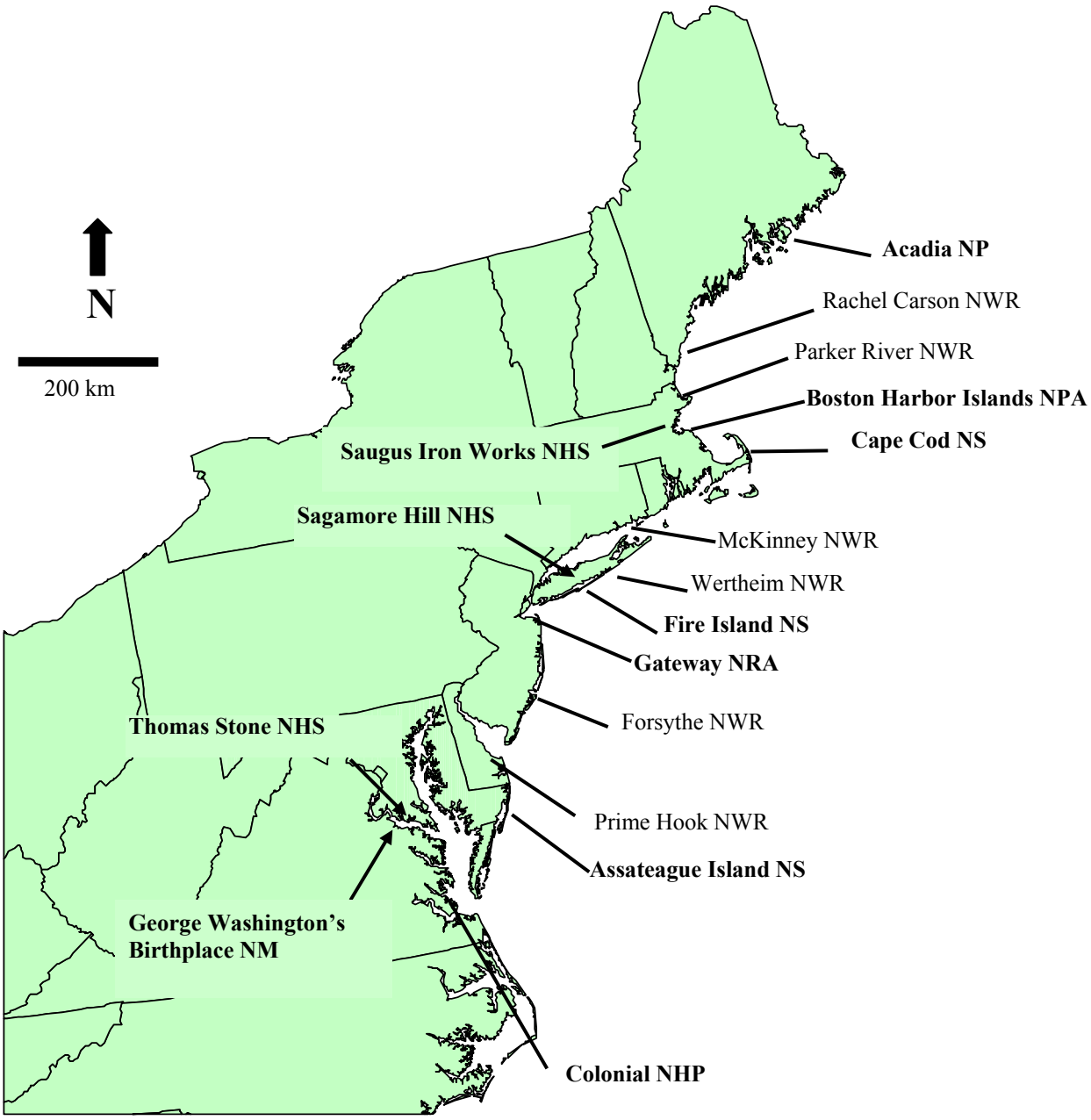


Figure 3. Northeast Coastal and Barrier and Northeast Temperate Network National Parks (in bold) and Region 5 US Fish and Wildlife Refuges where the Estuarine Nekton Monitoring Protocol is either currently implemented or will be implemented in the near future.

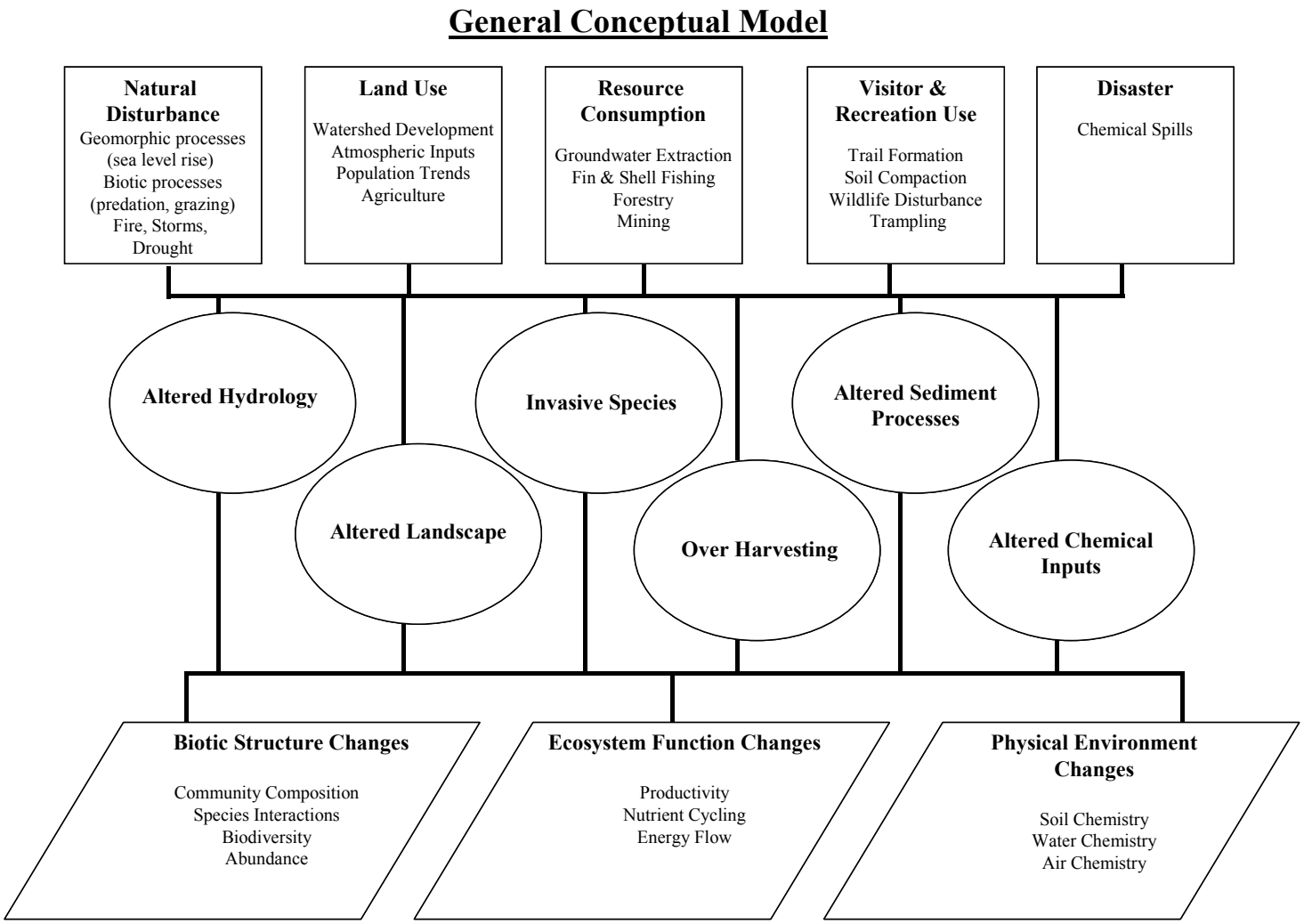


Figure 4. The Northeast Coastal and Barrier Network General Conceptual Ecosystem Model

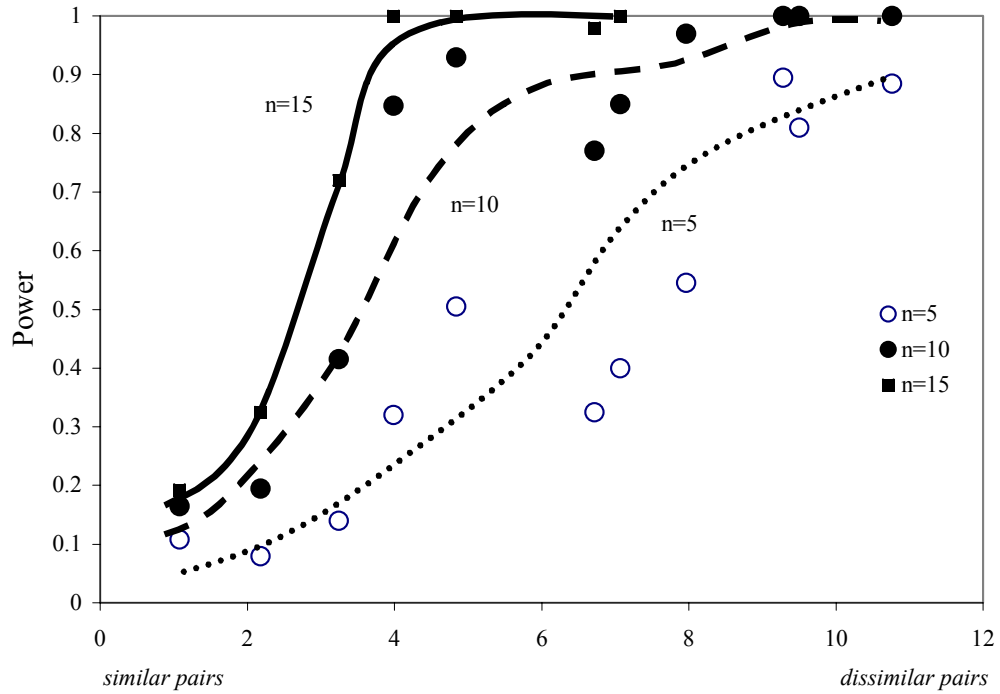


Figure 5. Power curves for sample sizes of 5, 10, and 15 with an alpha level of 0.05. Nekton density data from pairs of data sets that range in similarity from similar to dissimilar are compared.

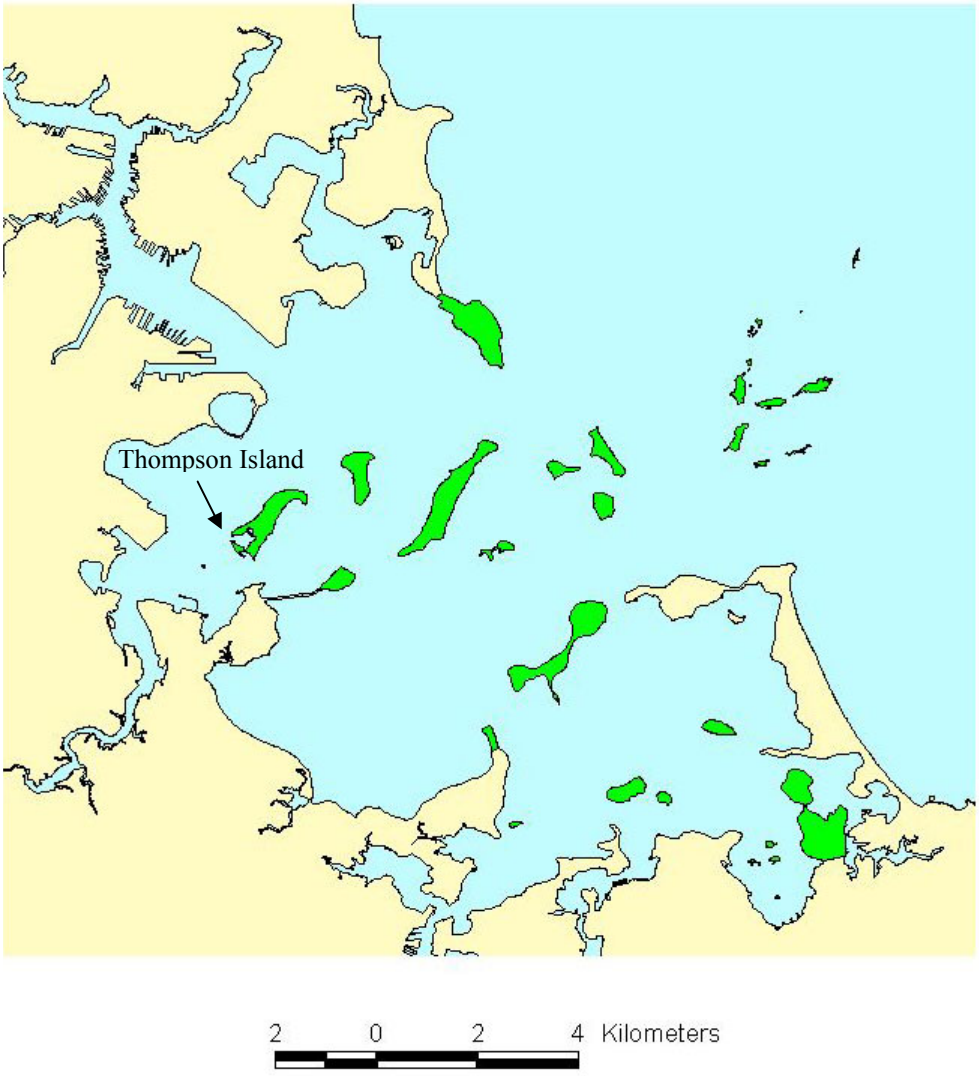


Figure 6. Map of BOHA showing sampling site.

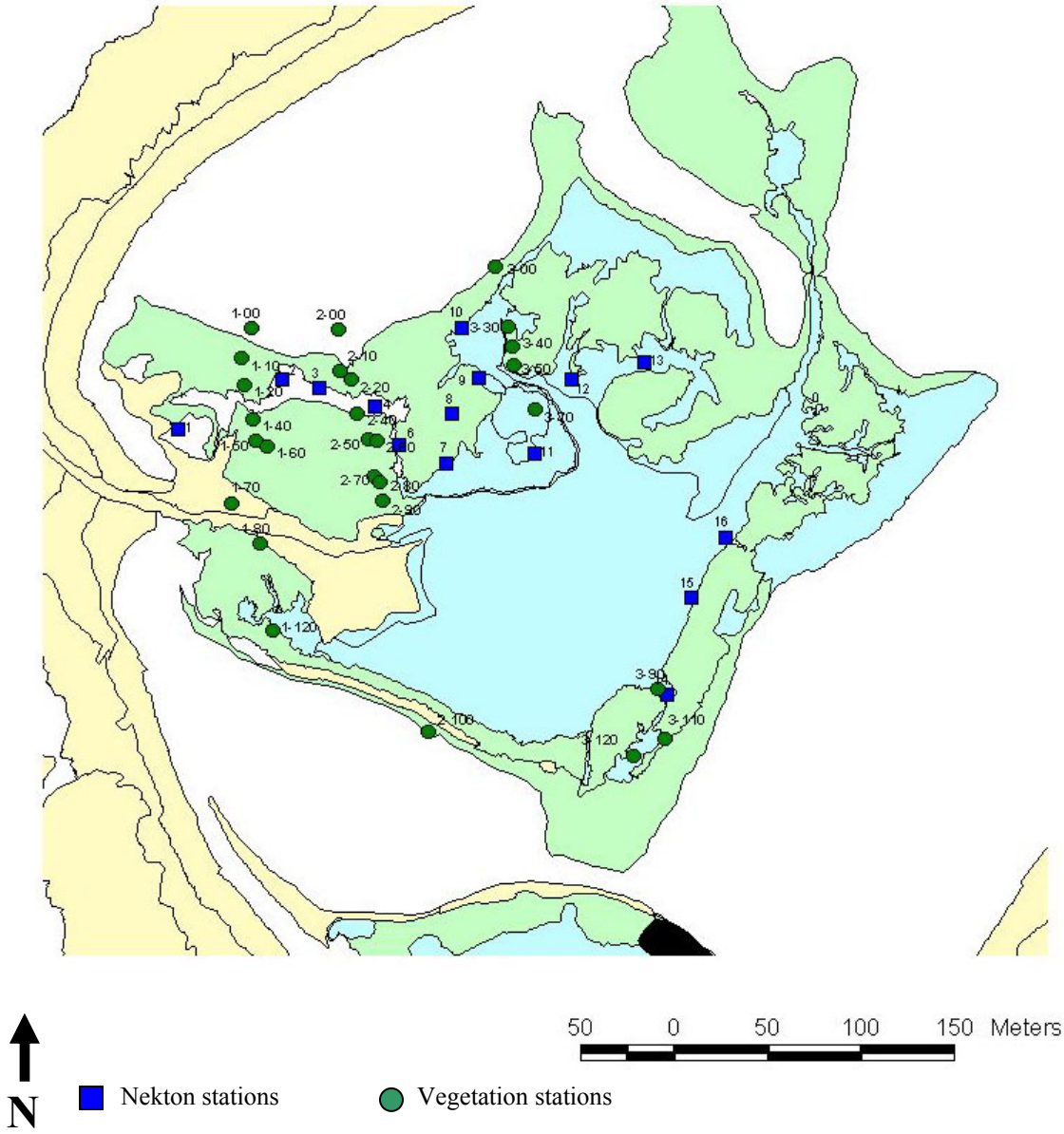


Figure 7. Map showing locations of stations sampled in 2004 for nekton and vegetation at Thompson Island, BOHA.

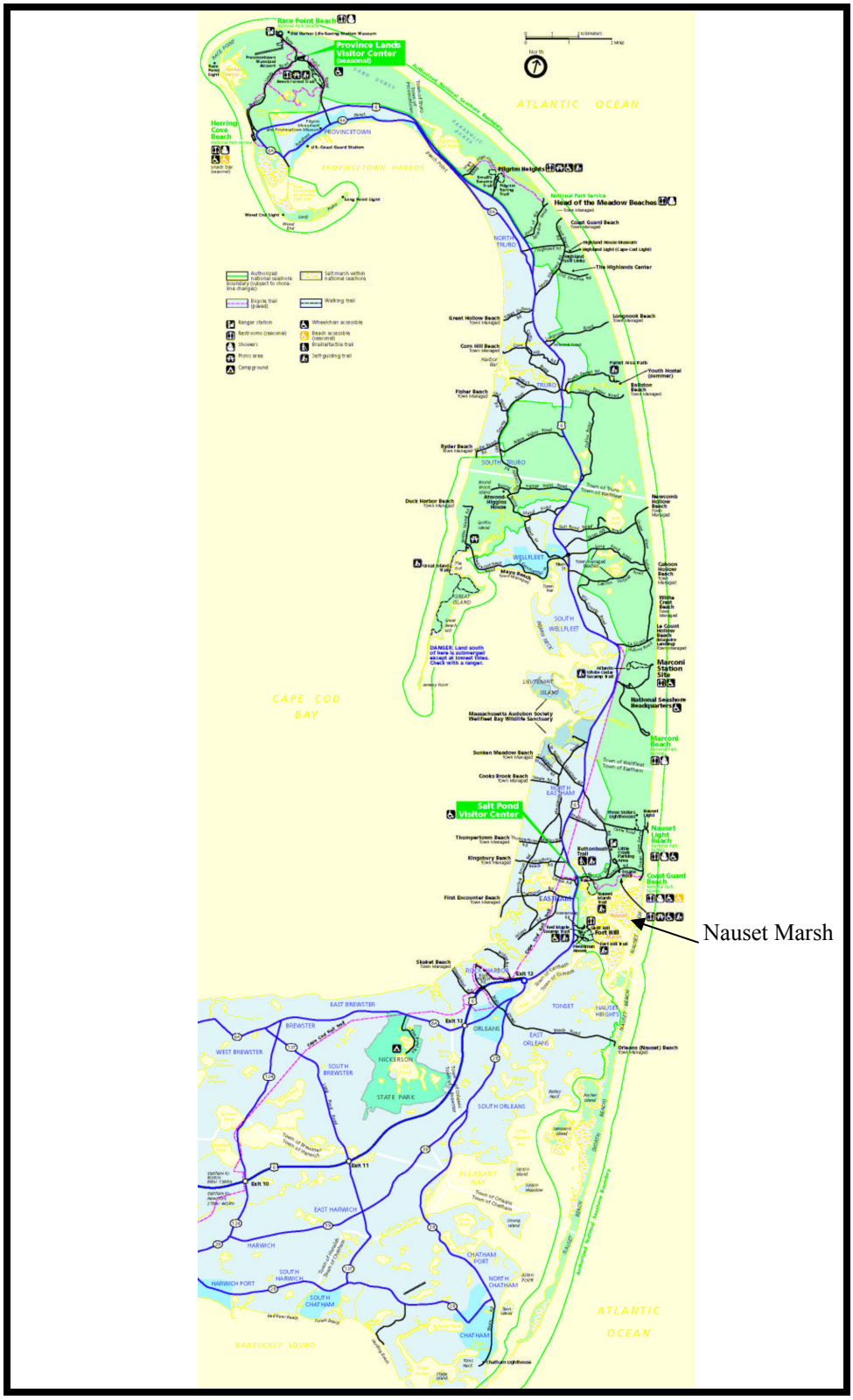


Figure 8. Map of CACO showing sampling site.

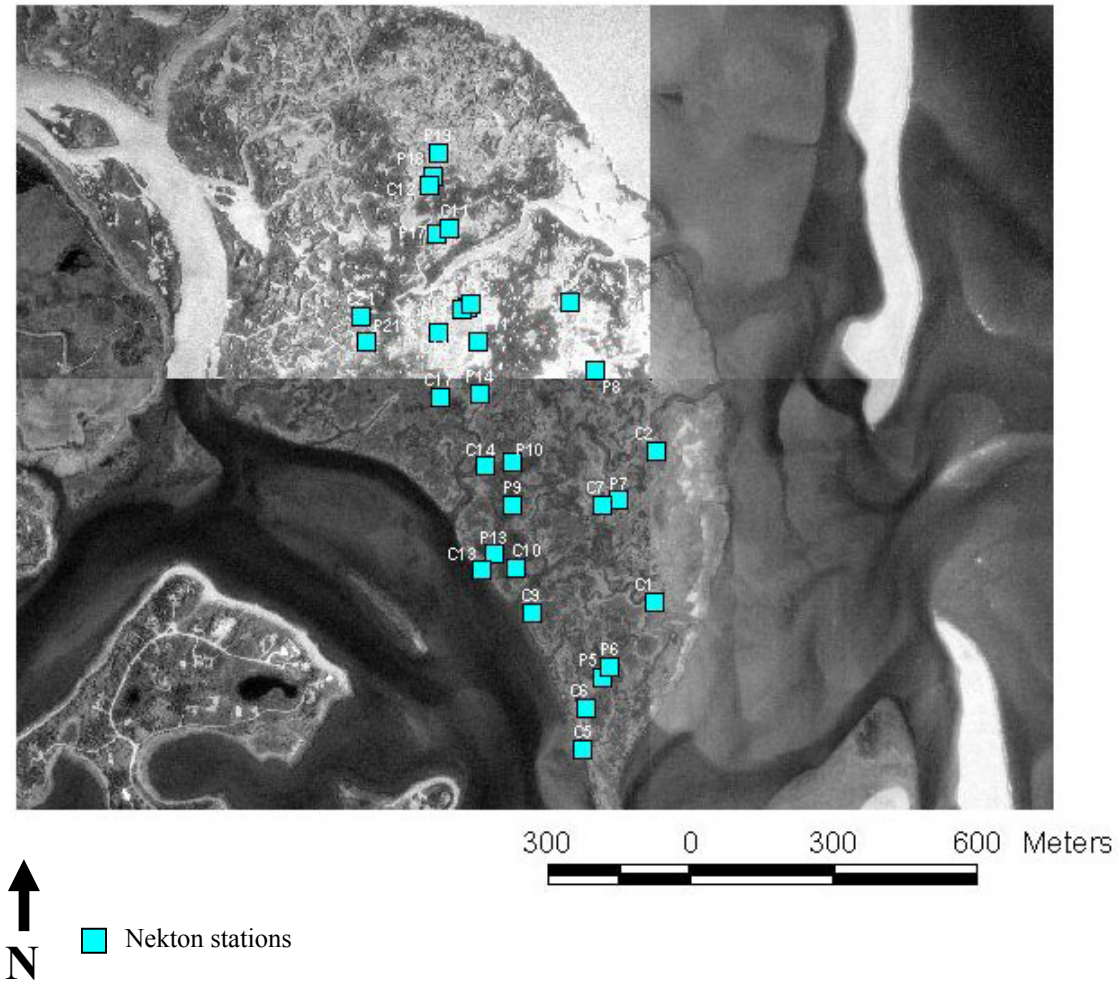


Figure 9. Map showing locations of stations sampled in 2004 for nekton at Nauset marsh, CACO. “C” indicates creek stations and “P” indicates pool station

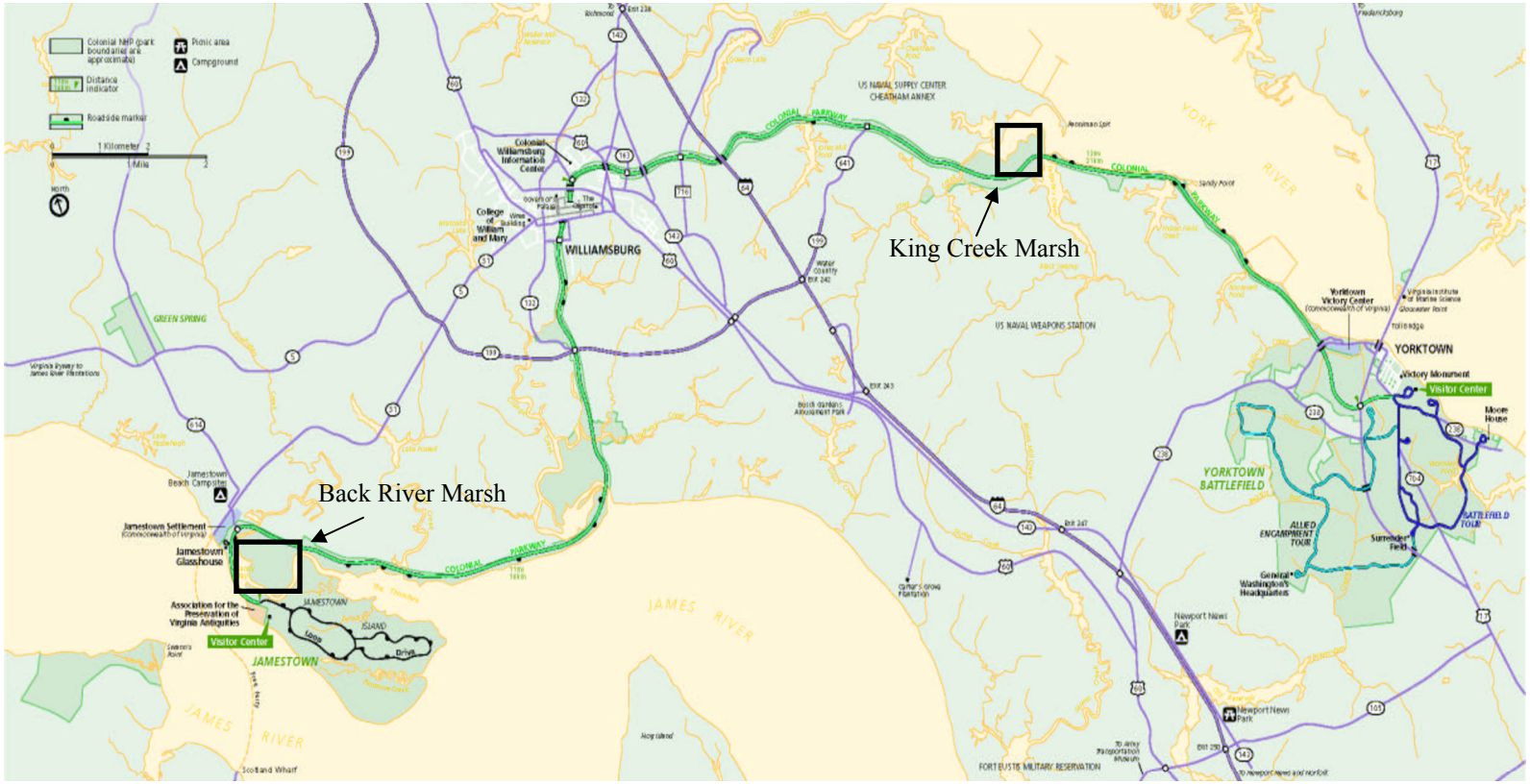


Figure 10. Map of COLO showing sampling sites.

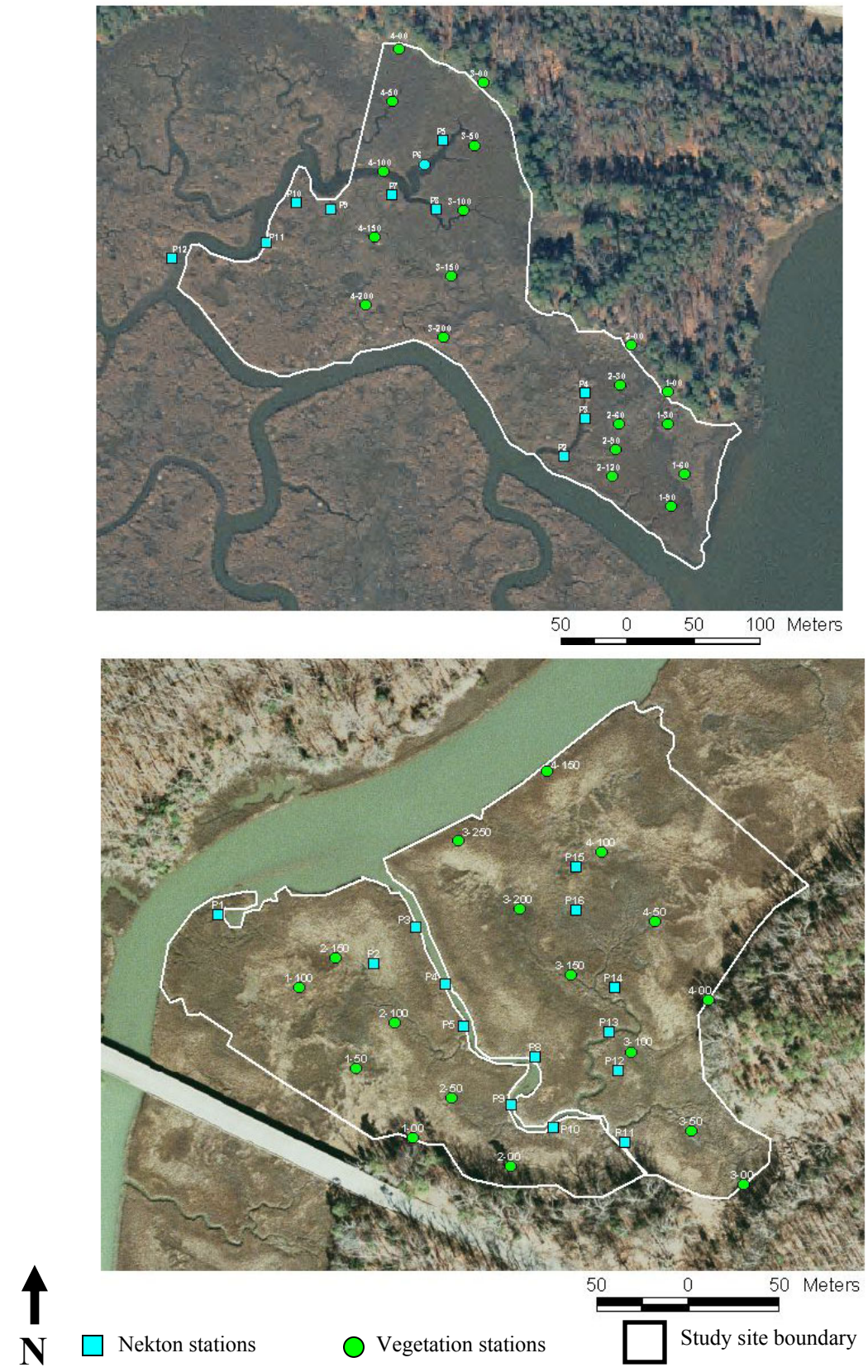


Figure 11. Map showing locations of stations sampled in 2003 for nekton and vegetation at Back River (top) and King Creek (bottom) marshes, COLO.

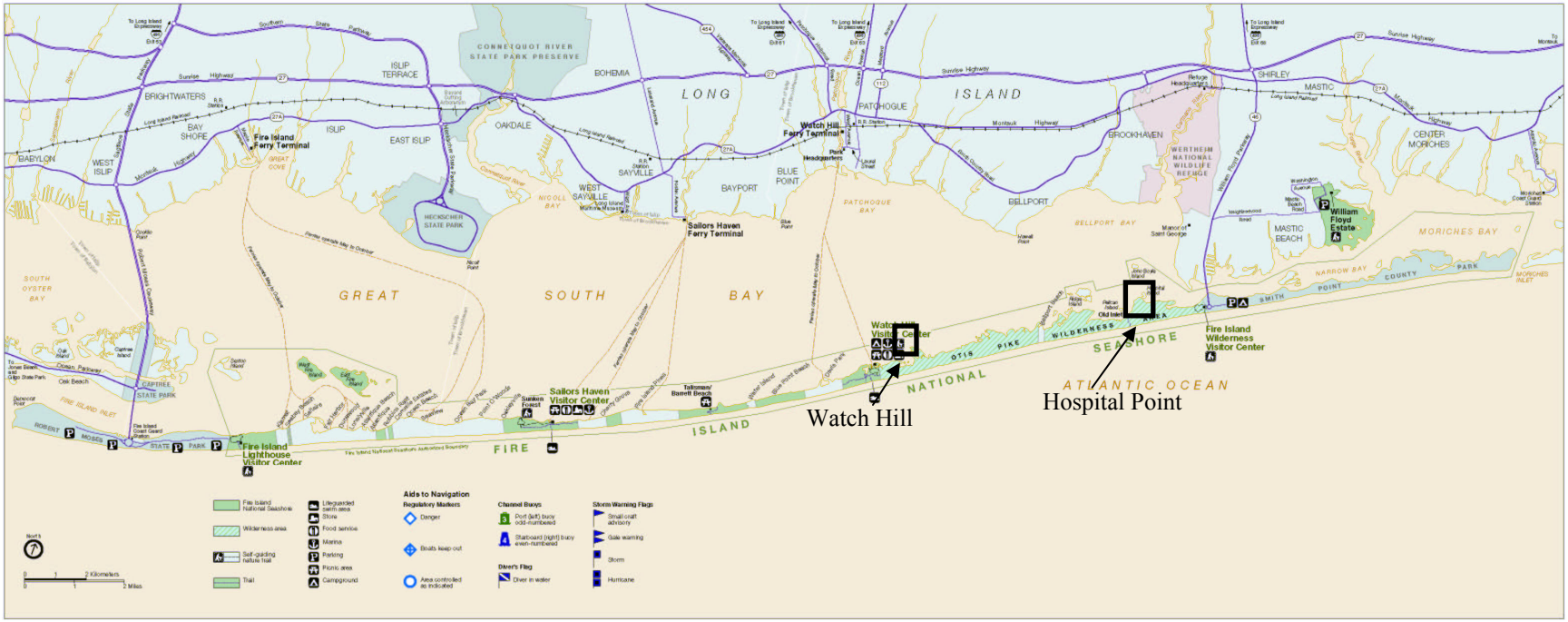


Figure 12. Map FIIS showing sampling sites.

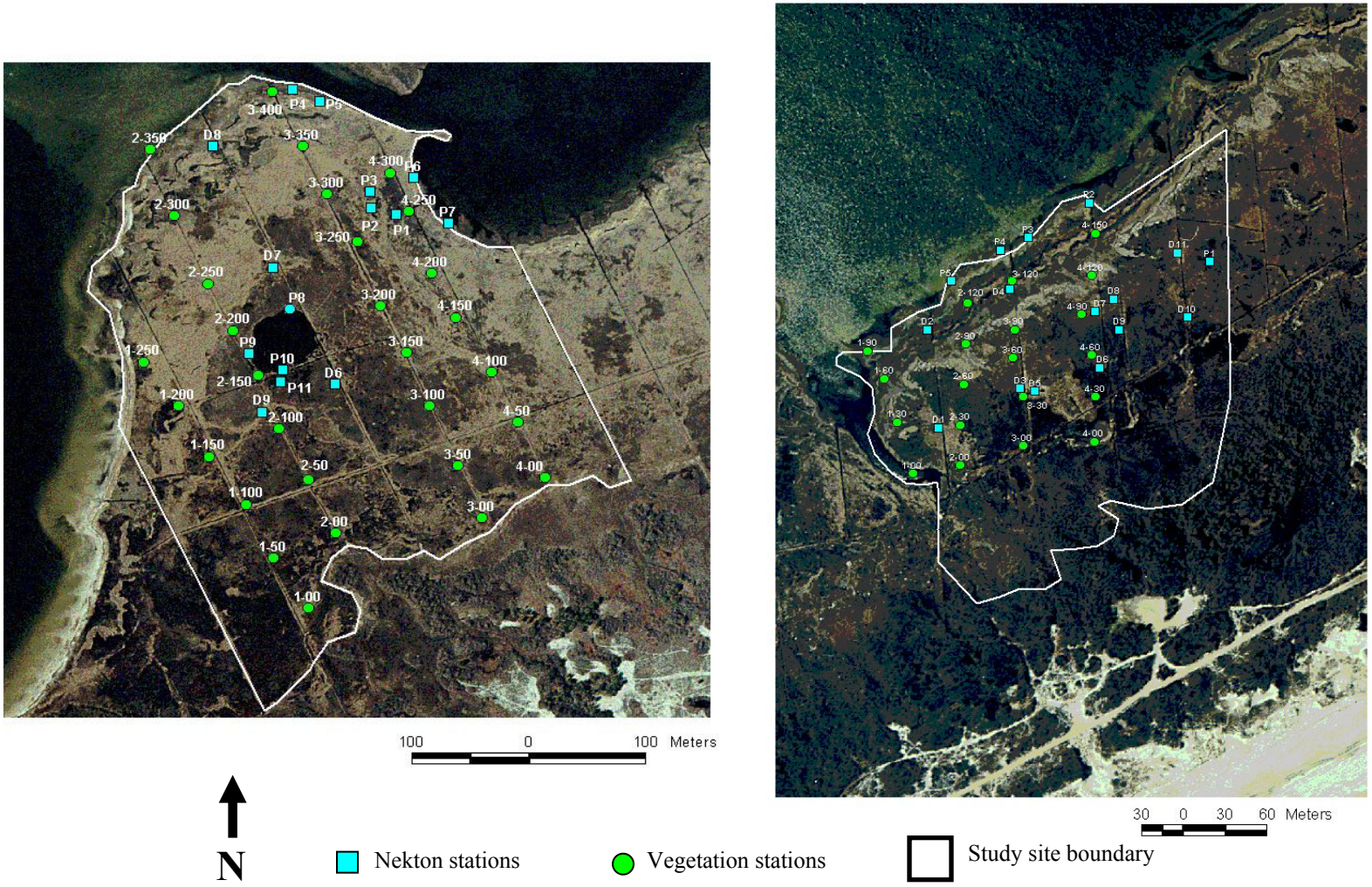


Figure 13. Map showing locations of stations sampled in 2003 for nekton and vegetation at Hospital Point (Left) and Watch Hill (right) marshes, FIIS. “D” indicates ditch stations and “P” indicates pool stations.

Figure14. Map of GATE showing sampling sites.

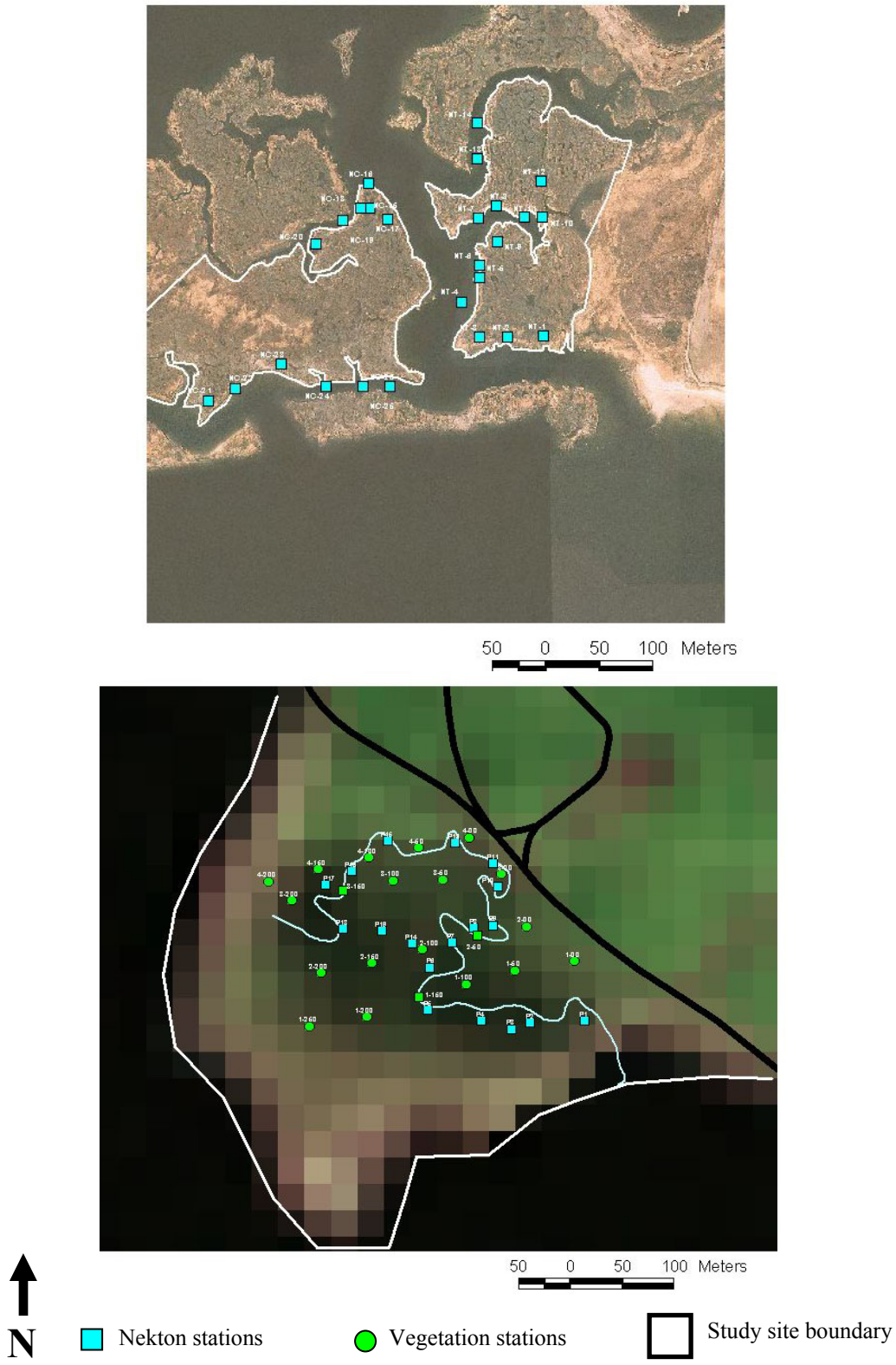


Figure 15. Map showing locations of nekton stations sampled in 2003 at Big Egg control and treatment marshes, Jamaica Bay Unit, (top) and Horseshoe Cove marsh, Sandy Hook Unit (bottom). (Note that station naming convention at Big Egg is different: NC=nekton control, NT=nekton Treatment.

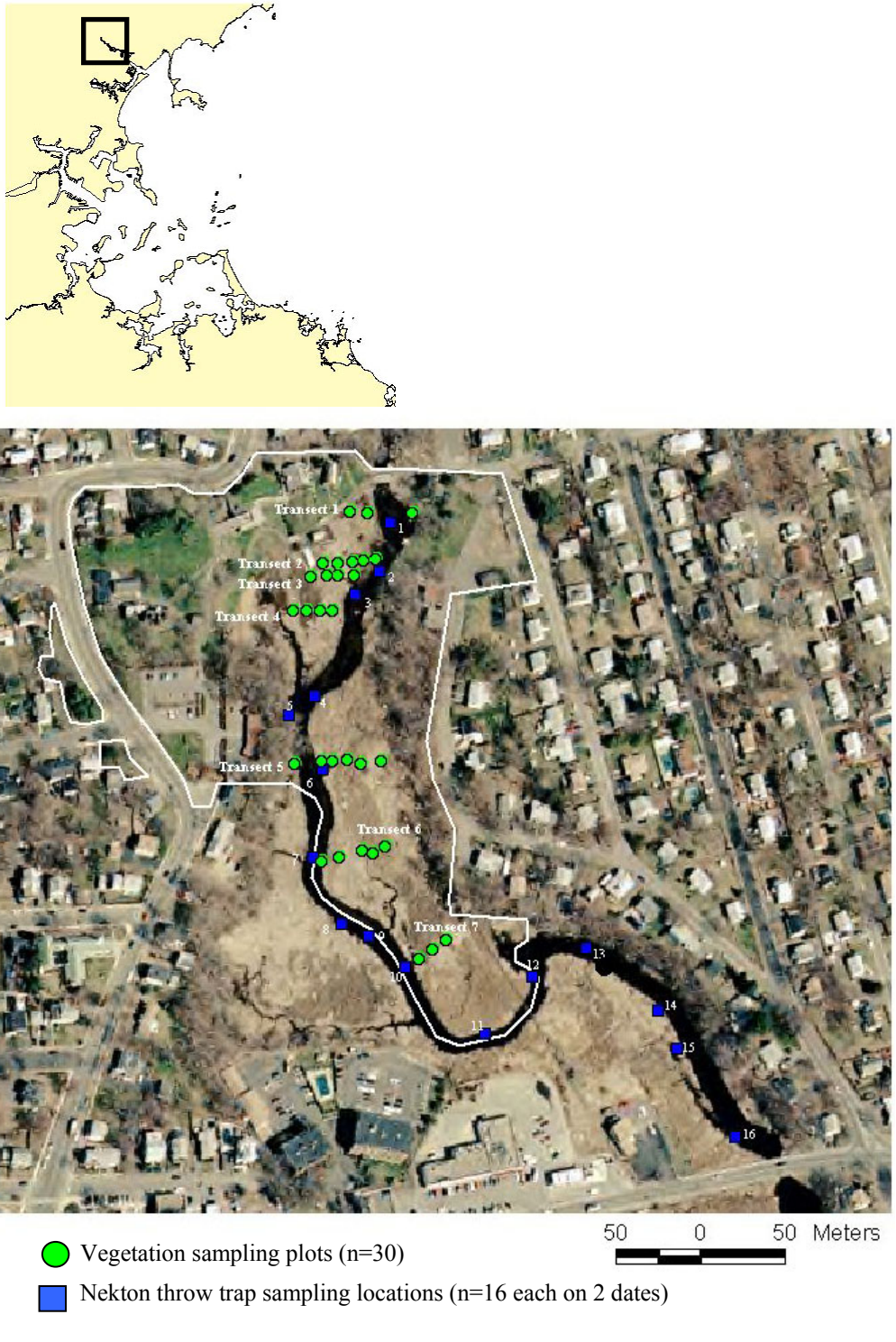


Figure 16. Map of Boston area (top) and map of SAIR Site (bottom) showing locations of nekton and vegetation stations sampled in 2004.

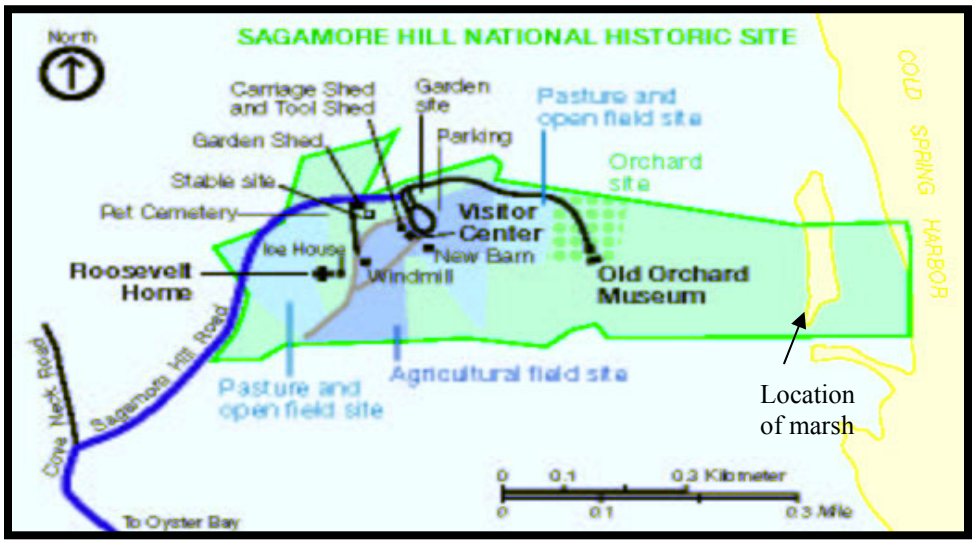


Figure 17. Map SAHI showing sampling site.

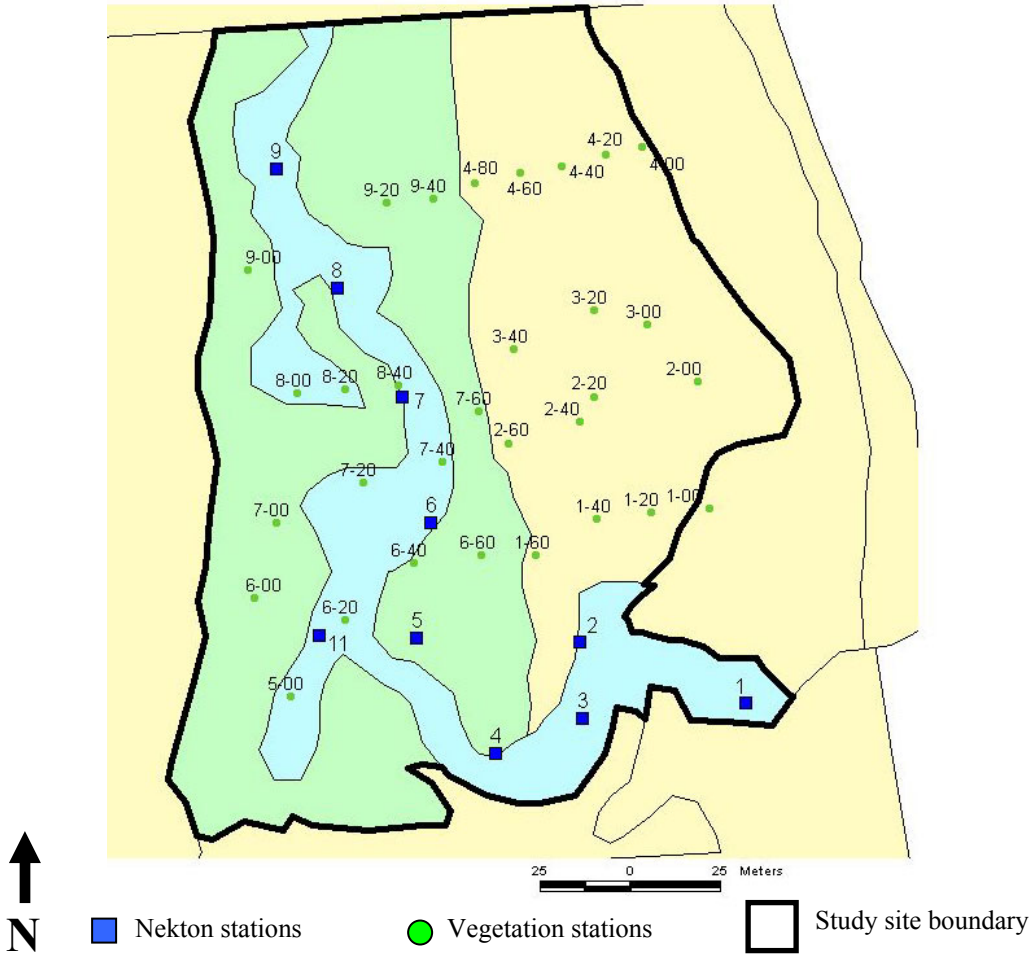


Figure 18. Map showing locations of stations sampled in 2004 at SAHI.

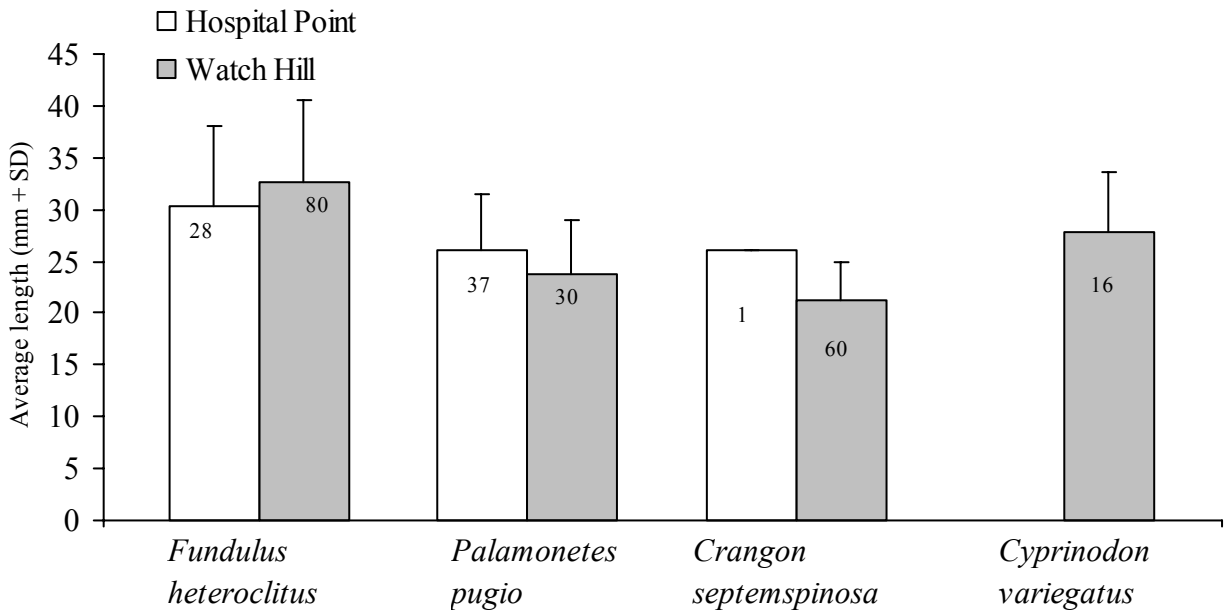


Figure 19. Average length (mm ±SD) of dominant nekton sampled at FIIS in 2003. Number of individuals measured indicated inside bars. Note: no *C. variegatus* were sampled at Hospital Point.

Standard Operating Procedures for Sampling Nekton in Salt Marshes

1 SOP 1: Selecting Study Sites

Study sites will be selected using a stratified random sampling design, if more than two sites are available within the park. If there are fewer than two salt marshes within the park, then either both areas will be monitored or one area will be randomly selected from the two areas.

- To stratify an area of extensive salt marsh, divide the area into equal sized strata such as distance from the inlet (*e.g.,* close, intermediate, and far from the inlet).
- Strata should be equal in size.
- Divide each strata into acceptable (*e.g.*, 1 to 7ha) areas.
- Randomly select a study area within each stratum from the available acceptable study areas.
- Considerations for acceptable study areas include:
  - There must be enough suitable habitat to sample. Since nekton is the target monitoring variable for this protocol there must be adequate habitat (*i.e.* marsh pools, creeks, mosquito ditches, shoreline area) to sample with the required number of replicates (15 to 50 stations) adequately spaced apart (at least 30m). If there is not enough habitat, for example if there are only a few pools, they can still be sampled, however, data maybe only be useful for species composition and not for comparing densities between sampling events
  - Access to study area
  - Co-location with existing monitoring programs
  - We have found that a size of 3ha to 8ha is a manageable study site area.

1.1 Existing Study Sites

1.1.1 Assateague Island National Seashore

- Moderate grazed marsh is located near Life of the Dunes Nature trail and is accessed from the nature trail parking lot. This area experiences moderate grazing pressure by the island's ungulates (*i.e.*, ponies). In the fall of 2005 SET's will be installed at this location.
- Valentines marsh is located in the southern end of the park near the Pirate Islands. This area experiences low grazing pressure by the island's ungulates (*i.e.*, ponies). In the fall of 2005 SET's will be installed at this location. This site must be accessed by 4-wheel drive vehicle via the beach.

1.1.2 Boston Harbor Islands National Park Area

- Thompson Island marsh is located on Thompson Island. Access to the marsh is by boat. Transportation is arranged through University of Massachusetts Boston, Division of Marine Operations (http://site.www.umb.edu/forum/1/Marine_Operations/res/web_site/index.html).

Vessel time was charged at a rate of $80 per hour in 2004. The landing craft is the best vessel for transportation as it can discharge passengers at the entrance of the marsh.

1.1.3 Cape Cod National Seashore

- Nauset Marsh is located within Nauset Estuary on the eastern side of Cape Cod. Access to the marsh is by boat. Boats (canoes and skiffs) are available through the Natural Resource Management Division at CACO.

1.1.4 Colonial National Historical Park

- Both marshes (Back River and King Creek) can be accessed from either public (King Creek) or National Park Service roads (Back River). Back River can also be accessed by canoe (obtained from the Natural Resource Management Division at COLO).

1.1.5 Fire Island National Seashore

- Both marshes (Hospital Point and Watch Hill) must be accessed by boat during the summer due to piping plover nesting on the back barrier beach which prevent access by 4-wheel drive vehicle. Boat transportation should be arranged (well in advance) through the Natural Resource Management Division at FIIS

1.1.6 Gateway National Recreational Area

- Big Egg Marshes (Jamaica Bay Unit) are accessed by boat. Boat transportation should be arranged (well in advance) through the Natural Resource Management Division at GATE.
- Horseshoe Cove Marsh (Sandy Hook Unit) is accessed via a public road adjacent to the marsh.

1.1.7 George Washington Birthplace National Monument

- Both marshes (Pope's Creek and Dancing Marsh) can be accessed from existing trails. The islands within Pope's Creek must be accessed by canoe.

1.1.8 Sagamore Hill National Historic Site

- The marsh at SAHI is accessed via a National Park Service nature trail (approximately 1km walk) to the marsh. A cart is available from the Natural Resource Management Division at SAHI, which makes carrying equipment to the marsh easier. The marsh is only partly owned by the NPS, the northern section (delineated by a chain link fence) is private property. Since the property owner has not given permission to sample on his property, sampling must only be on NPS property.

1.1.9 Saugus Iron Works National Historic Site

- Access to the marsh is by the parking lot in the maintenance area of SAIR. Natural Resource Management Division at SAIR will provide access to this locked area.

2 SOP 2: Establishing Nekton Sampling Stations

2.1 General Information for Establishing Nekton Sampling Stations

- This SOP describes methods for locating stations in marsh pools, tidal creeks, and nearshore areas.
- All sampling stations should be randomly selected prior to monitoring. There are a variety of ways to randomly select sampling stations and a few methods that could be used are described here. The most important thing to remember when locating sampling stations, regardless of the method used, is that the stations are selected RANDOMLY.
- Several methods are available to randomly select numbers (refer to following sections). Random number tables can be found in statistics textbooks, and random number generators can be found in spreadsheet software packages.
- Nekton sampling stations on ditches, tidal creeks, or shoreline areas should be at least 30m apart.
- There is no minimum distance for stations located on non-contiguous pools.
- Station locations on the same pool should be at least 30m apart or sampled more than 30min apart.
- Sampling station locations remain permanent for the sampling year, and from year to year are re-located using GPS or maps.

2.1 Sites with Fewer than 15 Pools

- If the study site has fewer than 15 pools than all pools should be sampled in the marsh in order to get the required replicate sample size (n=15) per sampling period.
- Pools can be sampled over a few days however all pools for a given sampling period and site should be sampled within 5 to 7 days.
- If a pool is large, more than one station may be located in it if additional stations are needed. However, stations located on the same pool should be at least 30m apart and should be sampled at least 30min apart.
- The exact sampling location within a pool is also randomly selected (refer to Section 1.5)

2.2 Sites with 15 to 100 Pools (approximately)

- If the study site has 15 to 100 pools (approximately), number all pools from 1 to the maximum number of pools.
- Determine how many pools can be sampled during each period based on the availability of staff and the required replicate size (n=15). For example, if 15 pools are be sampled, randomly choose 15 numbers between 1 and the maximum number pools on the marsh, these 15 numbers correspond to the pools that you will sample.
- An aerial photograph of your study site will easily allow the numbering of the pools and can be used as a guide to find the pools selected for sampling once you are in the field.

- The exact sampling location within a pool is also randomly selected (refer to Section 1.5)

2.3 Sites with more than 100 pools

Although a rare event, there is the possibility that a study site will have so many pools that it is impractical to number them using an aerial photograph. An example is Nauset Marsh within Cape Cod National Seashore. This marsh has hundreds of pools within the study area and thus numbering them in order to randomly select pools to sample is not logistically possible. If you are unable to number the pools from a map, then use either the transect method or the grid method (refer to section below) to randomly select pools for sampling. Both the transect and grid method are equally acceptable methods to randomly locate sampling stations at sites where there are many pools.

2.3.1 Transect method

This method uses randomly located transects and randomly located distances along transects as a guide to select pools for sampling. The stations along each transect are not expected to fall directly in a pool, instead they are meant to guide the sampler to a point within the marsh. Once at that point the person will sample the closest pool to that point. The number of transects is discretionary, but we suggest 4 to 6 transects per marsh. In the following example 6 transects each with 4 sampling stations (a total of 24 stations) were chosen (At Nauset Marsh we also sampled 24 creeks, thus our sample size was n=48). Using this method it is desirable that transects be more than 30m apart, thus this method is only appropriate for large marshes. This method is best performed in GIS, however, if good maps are available it can also be performed manually by hand-drawing on maps. The advantage of using GIS is that accurate distances can be measured and coordinates of points along transects where pools are to be sampled can be generated (using the calculate command in ArcView).

- Transects are randomly located by measuring one axis of the marsh and randomly selecting 6 numbers (the number of transects in this example) between 0 and the maximum distance of the marshes axis. These numbers correspond to the starting point of each transect.
- If the marsh has an elevation gradient (evident by vegetation patterns), transects should be oriented across this gradient (from low marsh or tidal creek to upland). If there is no gradient, then orientation of transects does not matter.
- We will use Nauset Marsh as an example to explain how this method is performed.
    - At Nauset Marsh, we also stratified our random transects within the marsh to ensure adequate coverage of the entire marsh area since the marsh was so wide (1000m). To accomplish this we divided the width of the marsh in to three 333m sections and randomly chose 2 numbers within each section (*i.e.*, 2 numbers between 1 and 333; 2 numbers between 334 and 666, and 2 numbers between 667 and 1000). Our 6 randomly selected

numbers for transect locations were 170m, 260m, 473m, 610m, 813m, and 921m (Fig. 2-1a).
    - Transects were drawn perpendicular to the marsh width at these distances.
- Transects are then drawn (in GIS or manually on a map) and their length is measured.
- Points along each transect that guide the sampler to a pool are also randomly located.
    - In this example we are using 4 points on each of the 6 transects for a total of 24 points (24 stations). Four random numbers are chosen for each transect. Each random number is between 1 and the length of the transect.
    - Since our transects were so long within Nauset Marsh, (950m to 1200m) we also stratified the points along the transect into 2 sections. Thus for the first transect (900m long), 2 random numbers were chosen between 0 and 450, and 2 random numbers between 451 and 900.
    - For example, on the first transect at Nauset (900m long) our sampling points for pools were 327m, 388m, 458m, and 688m (Fig. 2-1b).
    - By stratifying the points along the transect we are ensuring that the sampling points adequately represent the marsh area.
- Once the sampling points have been established, a map of the points and transects can be used as a guide to find pools in the field, or coordinates can be generated from a GIS program (*e.g.*, using the calculate command in ArcView).
- Once the sampler has located the point in the field, they select the closest pool for sampling. Since the point was randomly chosen, the closest pool to the point has also been randomly chosen.
- The exact sampling location within a pool is also randomly selected (refer to Section 1.5).

2.3.2 Grid method

The grid method uses a map of the sampling site that has been overlaid with a numbered grid. This method can be done in GIS or manually by overlaying transparent paper with a grid over the site map. An aerial photo or map of the site showing the location of all suitable habitat is necessary for either GIS-based or manual applications.

- Grid size can be arbitrary, but should be large enough that the number of grids is not overwhelming, but small enough to ensure that a random sample of the grids provides enough stations for sampling. We suggest grid size should be 1m to 10m square, depending on the size of the marsh.
- All grids that fall on a suitable habitat (i.e. pools) are numbered sequentially.
- 15 random numbers (the required replicate size) are then randomly selected from the total of numbered grids.
- The random numbers correspond to a numbered grid.
- The pool within the grid is sampled. If there is more than one pool in the grid, randomly select one pool to sample.
- Stations are located in the field.
- The exact sampling location within a pool is also randomly selected (refer to Section 1.5).

2.4 Location of Sampling Stations along a Pool's Perimeter

The specific location on the perimeter of a pool where the throw trap will be thrown from should be randomly located. We present two equally acceptable methods.

- A compass bearing between 0º and 360º is randomly selected and an imaginary line is drawn from the pool's center along the compass bearing, the intersection of the bearing with the pool's edge indicates the position of the station (Fig 2-2).

OR

- The perimeter of the pool is determined (by GIS or pacing) and a random number is selected between 1 and the pool's perimeter.
- The randomly selected number will indicate the distance from a point along the perimeter where the station will be located. The point along the pool's perimeter where the distance is measured from does not matter since the distance was randomly selected.

2.5 Locating stations on ditches, creeks, or shoreline areas

- To randomly locate sampling stations along ditches, creeks or shoreline areas, the distance of each ditch, creek or shoreline is measured.
- Random numbers between 1 and the total length of the ditch, creek or shoreline are then generated.
- The random numbers indicate the location of sampling stations along the creek or shoreline.
- Stations must be at least 30m apart, if they are not, a new random number should be generated, or one of the stations should be omitted.
- In marshes where there is an extensive creek network (that would prohibit accurate measurement of every single creek) the transect method (Section 1.4.1) or grid method (Section 1.4.2) can also be used to randomly select station locations.
- The grid method (Section 1.4.1) is also acceptable for locating stations along shoreline areas. In this case grids would be located along the shoreline and numbered.

2.6 Marking Sampling Stations

- Sampling stations should be located and marked in the field prior to sampling. If stations are located at the same time as sampling occurs, the nekton in the pool would have been disturbed by the activity associated with establishing the station and thus bias the subsequent sample.
- Stations are numbered sequentially from 1 to the total number of pools sampled.
    - Pools can be labeled "P" followed by the station number (*e.g.*, P1, P2).
    - Ditches can be labeled "D" followed by the station number (*e.g.*, D1, D2).
    - Creeks can be labeled "C" followed by the station number (*e.g.*, C1, C2).
    - This labeling scheme is more useful for keeping track of stations in the field rather than in the database, since the habitat sampled and gear used are always referenced in the database for each sampling station.
- Station locations should be clearly marked so that they can be re-located during sampling events.

- Oak stakes (1m in length) are a good marker, bio-degradable, and readily available from local hardware stores. Station numbers should be indicated on the oak stake with a permanent marker (which will need to be remarked every season) or burned into the wood (branded). Colored flagging can be attached to the stakes to aid in locating the stations.
- UTM coordinates of every station location should be recorded using a GPS.
- After GPS coordinates are taken, and before sampling begins, a GIS map should be plotted of the station locations to aid when sampling and to verify accuracy of station locations.

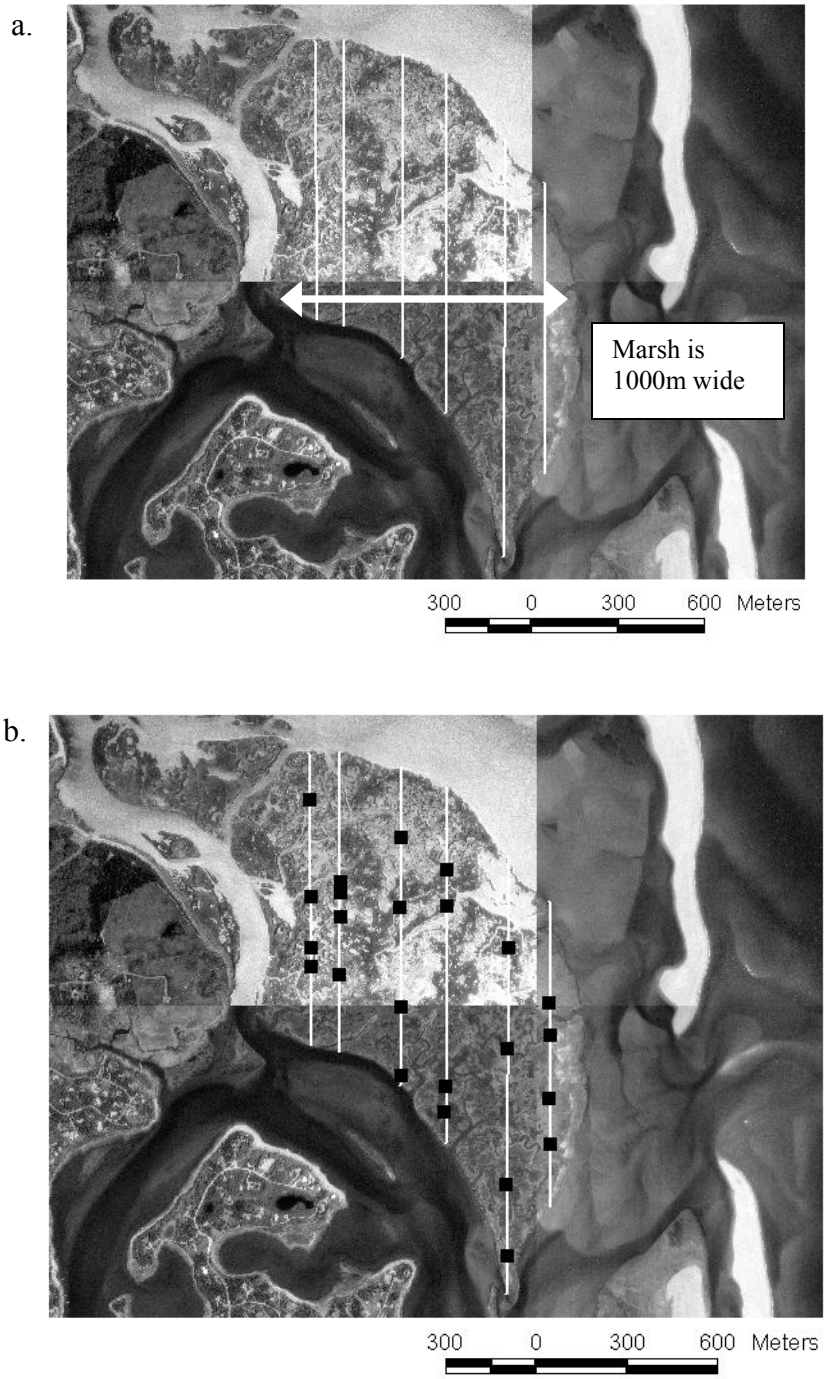


Figure 2-1. Photo of Nauset Marsh showing location of random transects (a) and random points (b) for locating sampling stations.

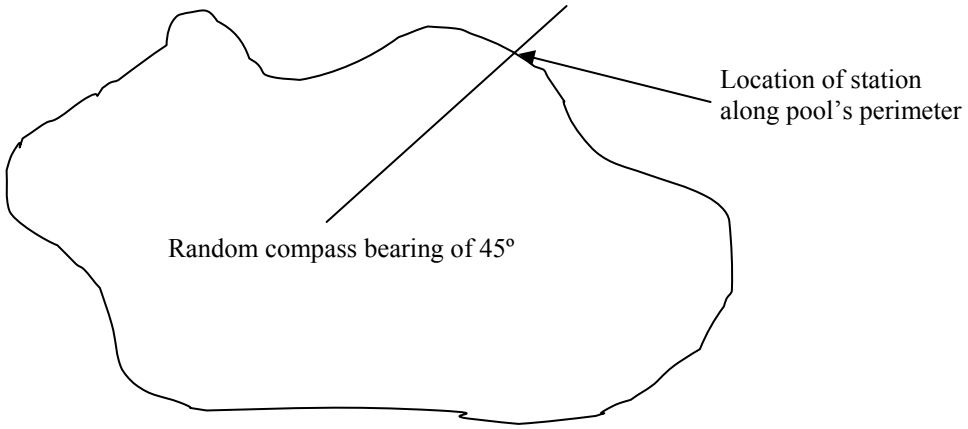


Figure 2-2. Diagram of random compass method to locate station along pond's perimeter.

3 SOP 3: Temporal Frequency of Sampling

3.1 Time of Year

- Nekton should be sampled twice per year, once in early summer (after June 15) and once in late summer-early fall (August to early October). Sampling prior to June 15 in the Northeast is not recommended because water temperatures are still cold and few nekton will be collected.
- The time frames for sampling nekton will vary due to differences in climates in the Network's region, for example nekton should be sampled later in early summer in Maine, whereas sampling as early as June 1 in Virginia is appropriate.
- The same sampling stations should be sampled during the two sampling events for each year.
- Stations should be re-located in subsequent years.

3.2 Time of Day

3.2.1 Diurnal Cycle

- Nekton should be sampled during daylight hours, unless specific data concerning nighttime densities are required.

3.2.2 Tidal Cycle

The timing of sampling depends on the tidal regime of the specific marsh and requires field reconnaissance to gather information on the flooding regime of the site, as sites will vary in the duration and amount of tidal flooding.

- Nekton sampled from pools should be sampled when the water has drained off the surface of the marsh (low or ebbing tide or prior to flood tide).
- Nekton sampled from tidal creeks or ditches should be sampled when water has drained off the surface of the marsh, but when there is still enough water in the ditches and creeks to sample (more than 10cm).
- Nekton sampling using a throw trap should occur at the same relative tide stage. To accomplish this, we suggest sampling in seaward habitats first (where the marsh surface drains earliest), and then proceeding to landward areas following the tidal prism. This method ensures that samples are collected at similar water depths throughout the marsh, and is thus one way to control for the effects of tide stage.
- Nekton sampling in ditches should occur at the same relative tide stage. Sampling salt marsh ditches should occur only after the marsh surface is drained of tidal water, but water still remains in the ditches. Sampling should occur on a high slack or ebb tide, when the marsh surface has drained. Timing the sampling for ditch nets is very critical, if the nets are set too late into an ebbing tide, the ditches will be drained before the nets are sampled. A thorough reconnaissance of the study site and its specific tidal regime should be well documented prior to ditch sampling.

3.3 Time Frame for Completing a Sampling Event

- All stations for a sampling event should take place within a 5-7 day period.

4 SOP 4: Field Crew and Training Procedures

4.1 Number of Staff

- One supervisor and at least 2 field technicians are the suggested number of staff to efficiently and accurately collect nekton monitoring data.
- A minimum of 2 people are necessary to physically sample nekton (for efficiency and safety in the field), but it is preferable to have more. A group of 4 people (2 teams of 2) dedicated to nekton sampling were used in the initial protocol testing phase.
- Two people are preferred to sample pools with throw traps. One to count and measure nekton and the other to record data.
- Two people are required to deploy the ditch net samplers.
- Since there are many replicates that must be sampled, it could take 1 team of 2 people 1-3 days to sample one marsh. As sampling must be closely coordinated with tides (sampling only after the marsh surface has drained during the daylight) a crew of 2 people could have a tight schedule to ensure that the samples are taken in a timely fashion.
- Individuals should be physically fit, be able to work long hours in field conditions, and able to carry the necessary equipment (*e.g.*, throw traps, ditch nets). Conditions in the field can be harsh so it is imperative that individuals conducting the sampling are able to tolerate typical summer conditions on a salt marsh (*e.g.*, extreme heat, mosquitoes, physical labor, extensive walking in hip boots).
- Throwing the throw trap takes practice and good upper body strength, and should be practiced before sampling in the field.

4.2 Training Procedures

- It is ideal for new staff to be trained by personnel who have previously sampled nekton using these protocols. Training should take place prior to the sampling season (*i.e.*, 1 to 2 weeks before the first scheduled sampling).
- A trial sampling trip should be conducted so staff can practice nekton identification and field sampling methods (*e.g.*, throwing the throw trap, deploying the ditch net) in the field.
- Staff should know how to use a GPS unit.
- Staff should be able to identify common nekton. They should be familiar with fish anatomy, terminology used in field guides (see Section 2.5.4), and common field identification characteristics of nekton. This can be learned on the job prior to sampling if trained by an expert in nekton identification.
- It is strongly urged that staff involve experts from local Universities or other agencies to assist with nekton identification.
- If voucher specimens are available from previous sampling, they should be studied by new staff.

4.3 Staff Qualifications

- A background in the sciences is preferred but not necessary.
- Familiarity with fishes is preferred, but not necessary.
- Individuals should be physically fit, be able to work long hours in field conditions, be able to carry the necessary equipment, and be able to meet travel and sampling constraints.

5 SOP 5*:* Field Season Preparation (Scheduling and Equipment Preparation)

5.1 Staffing Requirements

- A minimum of 2 people are necessary to sample nekton (for efficiency and safety in the field), but it is preferable to have more. A group of 4 people (2 teams of 2) dedicated to nekton sampling were used in the initial protocol testing phase. Since there are many replicates that must be sampled, it could take 1 team of 2 people 1-3 days to sample one marsh.
- As sampling must be closely coordinated with tides (sampling only after the marsh surface has drained during the daylight) a crew of 2 people could have a tight schedule to ensure that the samples are taken in a timely fashion.

5.2 Sampling Schedule

- A thorough reconnaissance of the study site and its specific tidal regime should be well documented prior to sampling. During this visit a map of the site should be taken into filed to verify the suitability of the sampling habitat (*i.e*., pools, ditches, creeks, *etc*.). Additional information on the duration of correct water conditions for sampling, especially for ditch nets, should be noted. At least one person who will be doing the actual field sampling should be present during this visit.
- Nekton should be sampled twice per year, once in early summer (after June 15) and in late summer-early fall (August to early October). Sampling prior to June 15, in the Northeast is not recommended because water temperatures are still cold and few nekton will be collected.
- The time frames for sampling nekton will vary due to differences in climates in the Network's region, for example nekton should be sampled later in early summer in Maine, whereas sampling as early as June 1 in Virginia may be appropriate.
- Sampling should occur after water has drained off the marsh surface. Nekton sampling must be closely coordinately with the specific tidal regime of each sampling site, which can make scheduling nekton sampling difficult as only two weeks per month will have tidal cycles where marshes are not flooded during daylight hours.
- Sampling for a specific period should be completed within 7 to 10 days.

5.3 Supplies and Equipment

The following supplies are required in the field to sample nekton:

5.3.1 Materials for Marking Station Locations

- Oak stakes or flags to mark station locations
- Mallet to pound stakes into ground
- Black permanent markers to mark transect and plot number on stakes
- Colored flagging (optional) to tie to oak stakes
- Compass
- Random number table (to determine specific station location at each pond)

- Aerial photos of study sites
- Draft map of study site showing boundaries of study areas and approximate location of ponds

5.3.2 Materials for 1m^2 Throw Trap and Dip Net Construction

- Drill, drill bits, saw, pliers, metal shears to drill and cut aluminum frame and hardware cloth of throw trap
- The throw trap measures 1m wide x 0.5m high. The bottom and top of the trap are open
- Eight, 1m long by 2.5cm aluminum bars
- Four, 0.5 m long by 2.5cm angle aluminum bars
- Nuts, bolts and lock washers to attach aluminum bars to angle bars
- 3mm hardware cloth (when reporting results from this method, investigators should cite a 3-mm mesh size, the mesh size of the throw trap)
- Thin gauge wire or cable ties to attach hardware cloth to aluminum frame
- Nylon netting, 3mm mesh, 4m long by 0.5m width (for skirt)
- 4m of float cord (for skirt)
- 1.3cm (1/2in) aluminum rod, approximately 4m long for dip net frame
- 1mm mesh nylon netting, 1.25m X 0.75m, for dip net

5.3.2.1 Throw Trap and Dip Net Fabrication

- The throw trap measures 1m^2 x 0.5m high.
- Construct the frame of the throw trap by attaching the 0.5m long 2.5cm angle aluminum angle bars (forms the corners of the trap) to the 1m long 2.5cm straight aluminum bars (forms the sides of the trap) with nuts, bolts, and lock-washers.
- Once the frame is built, the four sides of the trap are surrounded by 3mm mesh hardware cloth that is attached to the horizontal frame bars with thin gauge wire. Attach hardware cloth (with thin gauge wire or cable ties) to the 4 sides of the trap, leaving the top and bottom of the trap open.
- If water depths are expected to exceed 0.5m, the height of the trap can be extended to 1 m by attaching a skirt (3mm mesh nylon netting) to the top of the trap. The skirt is equipped with float-cord along the top edge to ensure that the top of the skirt floats at the waters surface.
- Bend the aluminum rod into the shape of the dip net (1m long by 0.5m wide) with a 0.5m handle.
- 0.5m length of 2.5-5.0cm diameter steel or PVC pipe can be fit over the aluminum rod handle of the dip net to strengthen the handle.
- Attach the 1mm mesh nylon net skirt (4m by 0.75m), to the dip net frame either with numerous small cable ties, or by sewing with twine or wire cable ties. Use of a 1mm mesh dip net facilitates collection of all nekton within the 1m^2 frame.
- When reporting results from this method, investigators should cite a 3mm mesh size, the mesh size of the hardware cloth.

5.3.3 Materials for Ditch Net Construction (for 1 net)

- Staple gun and staples, hog ringer gun and C-ring fastners
- Nylon netting (24lb test), 1/8in (3mm) mesh, at least 1m deep. Each net takes 5 meters of netting – a 1m X 3m section for the center of the net (sides & bottom) and two 1m X 1m sections the doors
- 20m of nylon rope, 3/16in (approx. 5mm) diameter. Each net takes 20m of line – four 4m lengths for rip cords and four 1m lengths for runner lines of the doors
- 5m of leadcore line; m for the top of each door (total 2m) and 3m for the floor of the net
- eye-hooks with 2.5cm eyes
- 4 oak stakes – 1.5 to 2m long, 2.5cm square
- Staple gun and 3/8 in stainless steel staples
- D-ring hand pliers and 9/16 in C-ring fasteners
- 25 to 30 plastic rings, rubber O rings, or links from plastic chain approximately 2.5cm diameter

5.3.3.1 Ditch Net Fabrication

- Cut a 1m by 3m section of the nylon netting for the center of the net.
- Cut two 1m by 1m sections of nylon netting for the doors of the net.
- Attach the doors of the net to the center section. The doors should be centered on the main body of the net along the 3m length (Fig 5-1a). To attach the doors take a 1m length of leadcore line and wrap the nylon netting from the leading edge of the door and the center 1m middle section of the net body around the leadcore and fasten the two pieces of nylon netting to the leadcore line with the D-ring pliers and 9/16 in C-ring fasteners.
- Attach 5 to 7 nylon rings or rubber O-rings to sides of the doors (side A in Fig 5-1a). Use the D-ring pliers to attach the rings to the nylon netting. The rings should be attached to the edge of the netting so the center of the ring is clear of the netting. The draw cord that pulls the doors up passes through these rings.
- Attach 3 to 5 plastic rings to the top of the door (side B in Fig. 5-1a). Use the D-ring pliers to attach the rings to the nylon netting. The rings should be attached to the edge of the netting so the center of the ring is clear of the netting.
- Attach a short length of lead core line to the top of each door (Fig. 5-1a, side B) using either cable ties or the D-ring pliers and C-ring fasteners. This is to weigh down the top of the net so it does not float up, and impede the passage of fish through the net.
- Attach a length (approximately 1m) of leadcore line to the bottom center of the net (Fig. 5-1b) on the outside of the net using either cable ties or the D-ring pliers and C-ring fasteners. This is to weigh down the center of the net so it does not float up when placed in the ditch.
- Attach the net to the four oak stakes using a staple gun and stainless steel staples. The free edges of the net (Fig 5-1a, side C, and Fig. 5-1b between points 1 and 2) are stapled to the oak stakes. The portion of the net closest to the doors should be stapled starting at approximately 30cm (1 ft) from the bottom of the oak stake, and continue up towards the top of the stake. The bottom 1ft of the stake should

be free of the net so that the stake can be pushed into the ground to hold the net in place while it is deployed.

- The runner lines are attached next. The runner lines hold the plastic rings close to the stake, so when the door is pulled up the net remains close to the stake.
- Attach the bottom of runner lines to the interior of the stakes (on top of the stapled netting). The runner lines are approximately 1m in length. The bottom of the runner line should be attached at the intersection of the doors and main body of the net. Tie a few knots in the end of the line and staple the line to the stakes using several staples close together on each side of the knot so the line will not pull loose.
- Pass the free end of the runner line through the 5 plastic rings that are attached to side A (Fig. 5-1a) of the door closest to the stake (Fig. 5-1b, runner line (3) and plastic rings (4)). The bottom most ring is added first, then the next ring, until all rings for that door side are on the runner line. The runner line is then pulled taut against the stake and the free end is stapled approximately 5 to 8cm above the end of the net. After stapling a knot should be tied in the free end of the line and stapled again on either side of the knot to ensure the runner line does not come loose.
- Attach the rip cord to the center ring on the top of the door, and pass the rip cord through one of the rings on the corner of the door. Then pass the rip cord through the top ring of the door that is attached to the runner line. Attach another rip cord to the same center ring, and pass it through the other corner ring, and the top ring on the other side of the door. When these lines are pulled, they will pull on the top rings attached to the doors, which in turn will pull the sides of the doors up the stakes to enclose the sides of the net.
- Attach the rip cords to the other side of the net as described above.
- Attach the eye-hook to the oak stake. When the net is held upright, with the 4 stakes sticking into the ground, the eye-hook should be placed on the outside of the stake. The free end of rip cord is passed through the eye-hook. When the rip is pulled the line should pass easily through the eye-hook, so the doors are pulled up smoothly.
- Label the stakes. We usually label the stakes A, B, C, and D. Be sure to label each net exactly the same. The labels are used to set the net correctly in the ditch and to measure the distance between the stakes in order to determine the area of the water that the net was fishing (refer to data sheet). For example, stakes A and B are place on one side of the ditch and stakes C and D are place on the opposite side of the ditch (Fig. 5-1b).
- Test each net to be sure that the rip cords pull up the doors smoothly and quickly.

5.3.4 Materials Needed for Sampling in the Field

- $1m^2$ throw trap and dip net
- Ditch nets
- Small ruler with mm increments (to measure nekton)
- Meter stick (to measure depth of water or ditch)
- Map of station locations

- Data sheets
- Pencils and permanent markers
- Identification keys
- Any other equipment necessary for taking environmental variables (*e.g.*, refractometer, oxygen probe, thermometer)

5.3.5 Personal Comfort and Safety Equipment in the Field

- Drinking water
- Hat
- Sunscreen
- Sunglasses
- Bug repellent and/or mosquito head netting
- Hip boots
- Snacks or lunch if sampling is for entire day
- Cellular phone or 2-way radio

We suggest that field staff inform either the supervisor or someone on the Park staff of where they will be sampling, what they will be doing, and an anticipated time of completion, so that in the case of an emergency the appropriate authorities can be informed of the location of the sampling crew.

5.4 Manuals and Identification Keys

We have found the following identification guides to be quite useful in the assisting with nekton identification. This is not an exhaustive list and staff are urged to draw upon local experts to assist with identification if necessary. There are also several websites that have extensive information on fish species. If voucher specimens are kept for later identification be sure they are retained in a fashion that preserves their characteristics. Nekton can be kept alive, and then later released to the same site where they were collected, or preserved in 70% ethanol (ETOH) after being humanly sacrificed.

Books:

Able, K.W. and M.P. Fahay. 1998. The First Year in the Life of Estuarine Fishes in the Middle Atlantic Bight. Rutgers University Press, New Brunswick, NJ,. ISBN# 0-8135-2500-4.

Bigelow, H. B. and W. C. Schroeder. 1953. *Fishes of the Gulf of Maine*. Fishery Bulletin of the Fish and Wildlife Service, Vol. 53, United States Government Printing Office, Washington, D. C.

Eddy, S. and J. C. Underhill. 1978. *How to Know the Freshwater Fishes*. Wm. C. Brown Company Publishers, Dubugue, IA. ISBN# 0-697-04750-4.

Gosner, K. L. 1978. *A Field Guide to the Atlantic Seashore*. Houghton Mifflin Company, Boston, MA. ISBN# 0-395-24379-3.

Raasch, M. S. 1997. *Delaware's Freshwater and Brackish-Water Fishes*. Delaware Nature Society, Dover, DE.

Robins, C. R., and G. C. Ray. 1986. *A Field Guide to the Atlantic Coast Fishes*. Houghton Mifflin Company, Boston, MA. ISBN# 0-395-39198-9.

Whitworth, W.R. *Freshwater Fishes of Connecticut*. 1996. State Geological and Natural History Survey of Connecticut, Department of Environmental Protection. Second Ed., Bulletin114, Hartford, CT. ISBN# 0-942081-08-0. (DEP Maps and Publication Office, 79 Elm St., Hartford, CT 06106, 806-424-3555).

Weiss, H. M. 1997. *Marine Animals of Southern New England and New York.* State Geological and Natural History of Connecticut Department of Environmental Protection, Bulletin115, Hartford, CT. ISBN# 0-942081-06-4.

Websites:

University of Wisconsin fish identification database:
http://mendota.limnology.wisc.edu/fishid/

FishBase- A Global Information System on Fishes**:** http://www.fishbase.org/home.htm

Fig. 5-1a

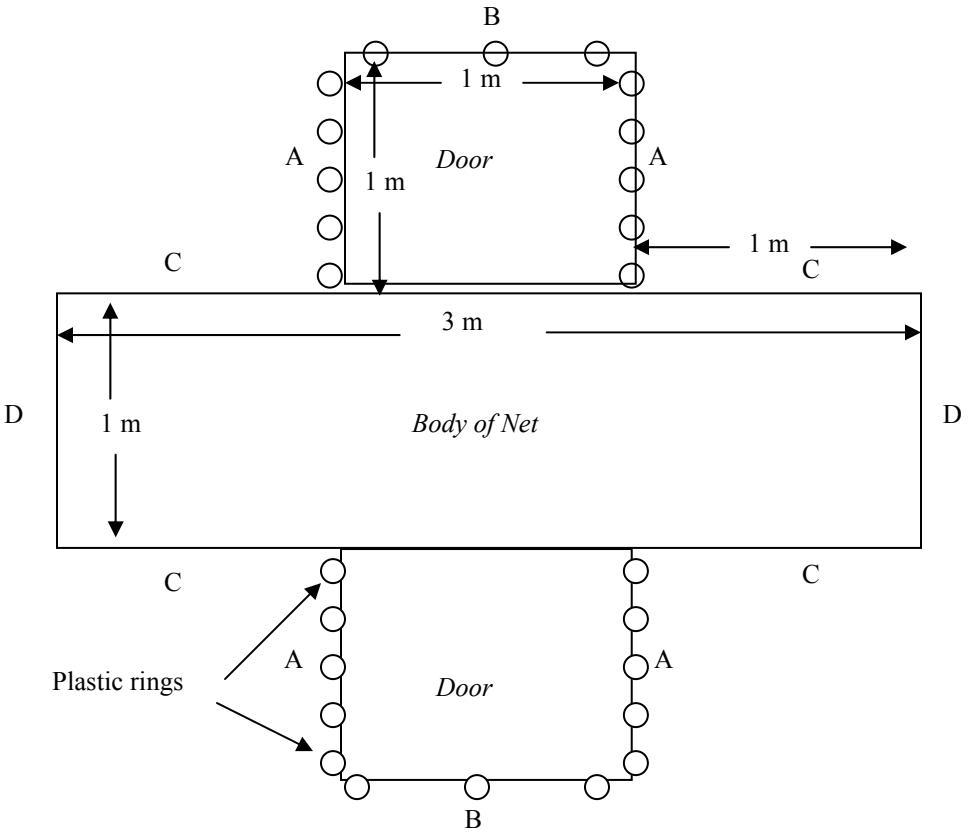


Fig. 5-1b

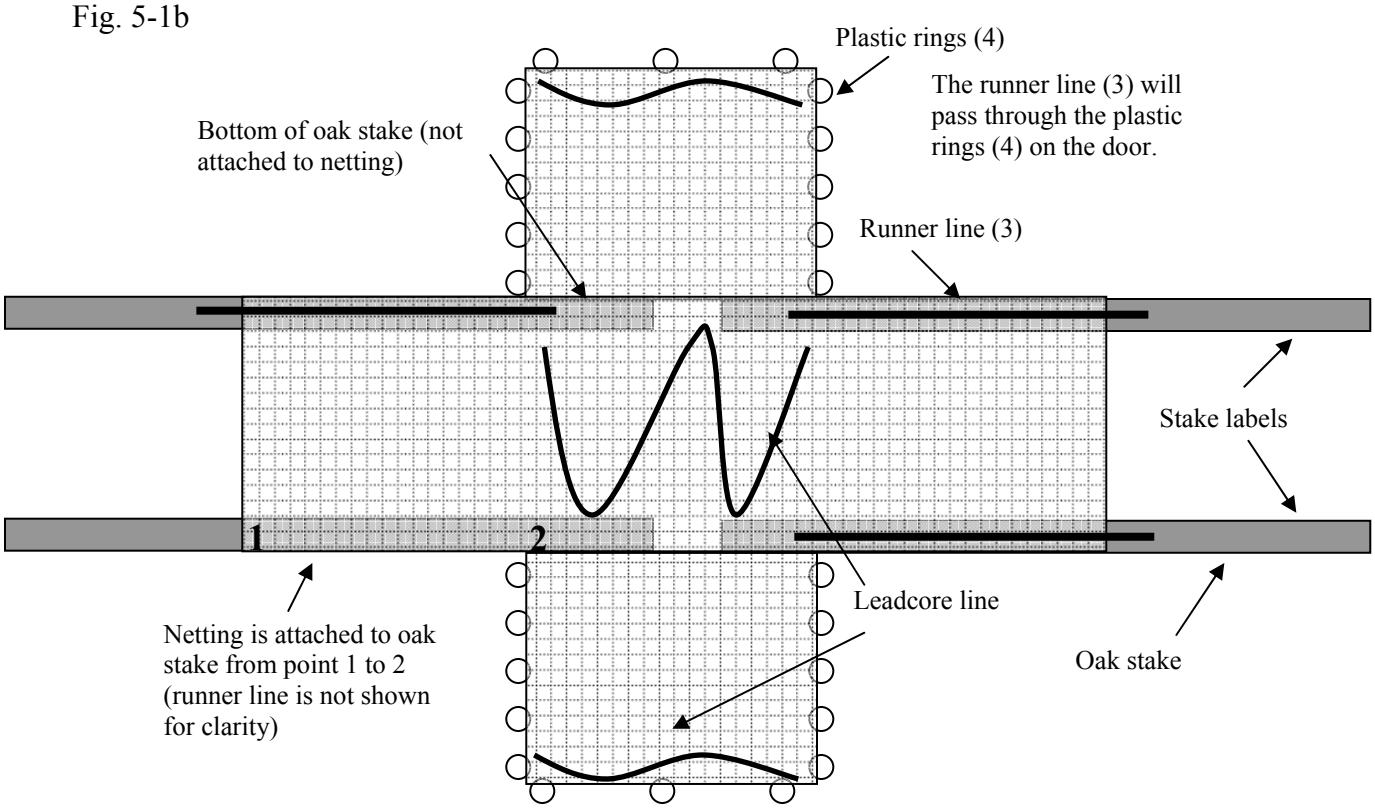


Figure 5-1a & b. Schematic of ditch net showing dimensions of nylon netting and attachment points for, plastic rings (5a), leadcore line, runner lines, and oak stakes (5b).

6 SOP 6: Using a GPS (placeholder for Network)

7 SOP 7: Sampling Procedures

Two sampling methods (throw trap [Fig. 7-1 & 7-2] and ditch net [Fig. 7-3]) are presented for sampling nekton. The preferred method is the throw trap (Fig. 7-1 & 7-2). With the throw trap, the species composition and abundance (density) of nekton (fish and crustaceans) is measured with a $1m^2$ enclosure trap in shallow water (< 1m) salt marsh habitats such as marsh ponds, tidal creeks, and shorelines.

The ditch net (Fig. 7-3) is a gear that is used for sampling small grid ditches. The throw trap is not a good sampling gear for the grid ditch habitat, as the trap is too large. Like the throw trap, the ditch net is also an enclosure sampling gear for sampling these narrow ditches. The center body of the net lines the sides and bottom of 1 linear meter (approximately) of ditch. There are two doors on the open ends of the net, which when pulled, rise up to close off the ends of the net, enclosing an area of water that is 1m long and as wide as the ditch. This sampling gear is designed to sample mosquito ditches and smaller tidal creeks up to 1m wide and 1m deep.

7.1 Data to be Recorded at Each Station

At each sampling station, regardless of the gear that is used, the following identifying information must be recorded on the data sheet (See Figs. 7-4 & 7-5).

- Date: Date of sample collection (month, day, year).
- Site: Name of park and study site.
- Station #: Station identification number. This should be a unique number for the sampling site.
- Sampling Crew: The initials the person or people conducting the sampling.
- Coordinates: The GPS coordinates of the sampling stations must be recorded. The preferred coordinate system is UTM, meters.
- Habitat Type: The appropriate habitat type should be circled on the data sheet.
- Percent cover of aquatic vegetation (pools only), if present.
- Time: Time of day the station was sampled.
- Temperature: water temperature in ºC.
- Salinity: salinity of water in ppt.
- Dissolved Oxygen: Dissolved oxygen of water in mgl^{-1}.
- Water Depth: Depth of water in pool, creek, or ditch in cm.
- Ditch Depth (ditch net only): Depth of ditch in cm.
- Tide: The appropriate tidal stage (ebb or flood) at time of sampling should be circled on the datasheet.
- Species: List each species that is collected. If common names are used in the field, the scientific names must be noted on the field data sheet as soon as possible to ensure accurate information is entered into the Access database.
- Tally: A tally of the number of individuals of a species that were collected, including the measured ones. This can be short hand notation (i.e., +10, +12, +36, +2, +5, *etc.*), as long as the total number (see below) is filled in upon returning to the lab.
- Total #: The total number of individuals of a species that were sampled. This can be filled in back in the laboratory if a calculator is required.

- Length: The length (in mm) of 15 individuals.

7.2 Sampling Procedure for Throw Trap

- Samples are collected by approaching to within 4 to 5m of a marked station with the throw trap.
- Approach the station quietly so as not to not disturb or startle the nekton. Only the person throwing the throw trap should approach the station, all others should remain at a distance (>10m) from the station to avoid startling the nekton.
- Pool stations are approached by crouching low and walking over the marsh surface, then waiting about 2 minutes before throwing the trap.
- There are two methods for throwing the throw trap depending on the physical ability of the person conducting the sampling.
    - Method 1: The trap is thrown into the water by tossing it from the hip like a giant Frisbee (Fig. 7-1). The trap is then quickly pushed into the sediment to prevent escape of nekton from under the trap.
    - Method 2: Throw the trap overhead (Fig. 7-2). This may be easier for those with less upper body strength or short arms. However, the distance covered by the trap is less using this method, and the sampler must stand closer to the station which is less desirable as nekton may be disturbed before the trap lands in the water.
- Repeat attempts (if the trap lands wrong) should be taken at least 30min apart.
- Once the sample is secured, nekton is removed by the large dip net.
    - The net is slid downward into the trap, flush against the side of the trap nearest the researcher.
    - The net is then moved across the trap with the forward edge of the net always remaining flush or slightly below the sediment until the opposite side of the trap is reached. In muddy sediments the dip net often goes through a thin layer of surface sediment, capturing buried nekton.
    - The net is then moved upward out of the trap, again keeping the leading edge flush against the far wall of the trap.
    - The dip net should be used from at least three sides of the trap because nekton may be hiding in the trap corners.
    - The dip-netting procedure is repeated until three consecutive dips do not capture any animals or if the first four dips come up empty. At this point the trap is considered empty.
- Ancillary environmental variables (water temperature, salinity, dissolved oxygen, water depth) should be measured at the time of collection.
- The surface area sampled for the throw trap is $1m^2$, therefore all density estimates for nekton sampled using a throw trap are number of nekton per m^2.

7.3 Sampling Procedure for Ditch Net

7.3.1 Deploying ditch nets

- Nets are placed at the station locations in the ditches at least 30min before sampling. This usually means that the nets are placed at flood or slack tide.

- To set up a ditch net requires 2 people, each standing on opposite sides of the ditch.
    - One person will take stakes labeled “A” and “B” and place the stakes into the bottom of the ditch close to the side of the ditch.
    - The other person will take stakes labeled “C” and “D” and place them on the opposite side of the ditch.
    - The net should be stretched tight between stakes “A” and “B” and stakes “C” and “D” so that approximately a 1m section of ditch is sampled. (Fig. 7-3).
- The lines from the doors should be pulled to make sure that the lines are not fouled and that the doors will pull up smoothly and quickly.
- Push the doors and the center of the net down into the bottom of the ditch with the meter stick. Make sure that the net lays down on the bottom of the ditch, so that fish passage through the net is not impeded.
- Measure the distance between all the stakes (*e.g.,* “A” to “B”, “B” to “C”, “C” to “D”, and “D” to “A”) and the diagonal distance between stakes “A” and “C” and record these on the datasheet. These distances are measured when the net is placed in the ditch and are necessary to calculate the area of water (sum of 2 irregular triangles) that is sampled.
- Lay the lines from the doors out on the marsh surface as far from the net as possible without pulling on the doors.
- Note the time that the net is deployed on the data sheet.

7.3.2 Sampling ditch nets

- Ditch nets should not be sampled until they have been deployed for at least 30min. This time period is necessary to minimize any disturbance to nekton caused by placing the net in the ditch.
- Ditch nets are sampled at high slack or ebb tide.
- Two people are required to pull the ditch nets.
- The nets are quietly approached from opposite sides of the ditch, one person on each side.
- Upon reaching the lines from the doors, each person kneels and waits quietly for approximately 2min. The lines to the doors should not be handled during this time, as the vibrations on the lines can be transmitted to the stakes and possibly disturb nekton that are in the net. At a pre-determined signal, both people quickly pull on the lines and run towards the net. The doors of the net will pull up, enclosing nekton within the net (Fig. 7-3).
- The net is then quickly lifted out of the ditch and onto the marsh surface. The best way to do this is to have both people pull the stakes out simultaneously (while still maintaining pressure on the lines from the doors).
- All four stakes are then handed to one person who will lift the net out of the ditch and onto the marsh surface. It is important to quickly pull the stakes and net out of the ditch, since this creates a bag of netting in the center of the net where the fish are trapped.

- The net is then laid out on the marsh surface and the nekton are identified, counted, and measured.
- The collection time is recorded.
- Ancillary environmental variables (water temperature, salinity, dissolved oxygen, water depth, creek depth) should be measured at the time of collection.
- The surface area sampled from a ditch net is calculated from the sum of two irregular triangles. Figure 6-6, is an example of how this calculation is performed.
- Density estimates for nekton sampled using a ditch net are presented as number of nekton per m^2.

7.4 Processing the sample

- In each sample, up to fifteen individuals of every species are measured to the nearest mm for total length (from the tip of the snout to the tip of the caudal fin for fishes; from the tip of the rostrum to the tip of the telson for shrimp) or carapace width for crabs (the distance between the two furthest points across the carapace).
- Nekton may be identified using any number of guides (refer to Section 4.4: Manuals and Identification Keys).
- Individuals that are difficult to identify and voucher specimens should be humanely sacrificed by a strong blow to the head, preserved in 70% ethanol (ETOH), and returned to the laboratory for identification. All voucher specimens should be stored in appropriate containers and clearly labeled with the contents (type of preservative), species, date, site, and station number.

7.5 What to do if the Station is Dry

Stations can be sampled with a throw trap as shallow as a few cm in depth. But, we suggest that if there are stations that may potentially go dry during the summer in between sampling periods (*e.g.*, shallow salt pannes), that additional stations should be added to prior to the beginning of the season to compensate for the possibility of dry stations during the late summer sampling period. Ditch nets should be sampled when there is 10cm or greater water depth.

- Occasionally, a station set up prior to sampling will be dry when it comes time to sample. If the sampling station is dry, and it is the first round of sampling, simply randomly relocate the station to another suitable habitat where there is water.
- If the station has previously been sampled in an earlier round of nekton sampling, simply note that the station was dry and no data were taken. This will decrease the number of replicates for this sampling period, however, since the same stations should be sampled during each sampling period, we advise against moving a station location in the midst of a sampling season.

7.6 Data Sheets

An example of a throw trap and ditch net data sheet used to record sampling events are shown in Fig. 7-4 and 7-5, respectively.

- All information should be filled out on the data sheets in the field.

- If species are identified back in the lab, the person verifying the identification should date and initial the identification.
- Any changes or edits to information on the field data sheet must include the date and initials of the person making the change.
- Upon return from sampling, all data sheets should be checked to make sure they include all information. If any information is missing every attempt should be made to complete the missing information. The person completing the missing information must initial and date the change and/or addition.

Figure 7-1. Sampling technique for $1m^2$ throw trap. The trap is tossed like a frisbee into the pond that is being sampled.

Figure 7-2. Overhead method for throwing $1m^2$ throw trap.

Figure 7-3. Photos of ditch net in the field showing correct deployment (top), doors being pulled up (middle), and the net once the doors have been pulled (bottom).

Throw Trap Data Sheet

SITE:_____ DATE: _____

STATION #:_____ SAMPLING CREW: _____

Coordinates: N: _____ E:_____

Habitat Type: Pool/Panne Tidal Creek Plugged Ditch Open Ditch

Aquatic Veg. Species & cover (if present): _____
(cover classes: <1% 1-5% 5-10% 11-25% 26-50% 51-75% >75%)

Time:_____

Water temp: _____ Salinity: _____ DO: _____

Water Depth: _____ Tide: Flood or Ebb

NEKTON SPECIES & MEASUREMENTS

SPECIES #1 _____ Total # of individuals: _____

Talley (include measured fish): _____

LENGTHS (15): _____

SPECIES #2 _____ Total # of individuals: _____

Talley (include measured fish): _____

LENGTHS (15): _____

SPECIES #3 _____ Total # of individuals: _____

Talley (include measured fish): _____

LENGTHS (15): _____

SPECIES #4 _____ Total # of individuals: _____

Talley (include measured fish): _____

LENGTHS (15): _____

SPECIES #5 _____ Total # of individuals: _____

Talley (include measured fish): _____

LENGTHS (15): _____

Figure 7-4. Throw trap data sheet

Nekton Ditch Sampler Data Sheet

SITE:_____ DATE: _____

STATION #:_____ SAMPLING CREW: _____

Coordinates N: _____ W: _____

Habitat Type: Tidal Creek Open Ditch Plugged Ditch

Deployment Time: _____ Collection Time: _____

Distance: A to B:____ B to C:____ C to D: ____ D to A: ____ Diagonal _____

Water temp: _____ Salinity: _____ DO: _____

Water Depth: _____ Creek/Ditch Depth: _____ Tide: Flood or Ebb

NEKTON SPECIES & MEASUREMENTS

SPECIES #1 _____

Talley: _____ Total # of individuals: _____

LENGTHS (15): _____

SPECIES #2 _____

Talley: _____ Total # of individuals: _____

LENGTHS (15): _____

SPECIES #3 _____

Talley: _____ Total # of individuals: _____

LENGTHS (15): _____

SPECIES #4 _____

Talley: _____ Total # of individuals: _____

LENGTHS (15): _____

SPECIES #5 _____

Talley: _____ Total # of individuals: _____

LENGTHS (15): _____

Figure 7-5. Ditch net data sheet

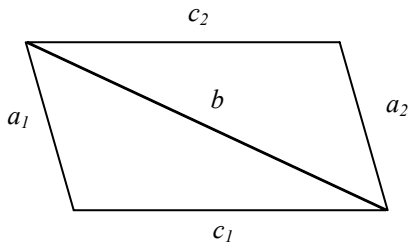
Calculating the Area of a Ditch Net

The area of a ditch net is calculated as the sum of two irregular triangles. The areas of the 2 irregular triangles are calculated from the 5 distances measured in the field.

$$\text{Area sampled } (m^2) = \sqrt{[s_1 * (s_1 - a_1)(s_1 - b)(s_1 - c_1)]} + \sqrt{[s_2 * (s_2 - a_2)(s_2 - b)(s_2 - c_2)]}$$

Where:

a_1 = side one of triangle 1
c_1 = side two of triangle 1
b = diagonal between triangle 1 and 2
a_2 = side one of triangle 2
c_2 = side two of triangle 2

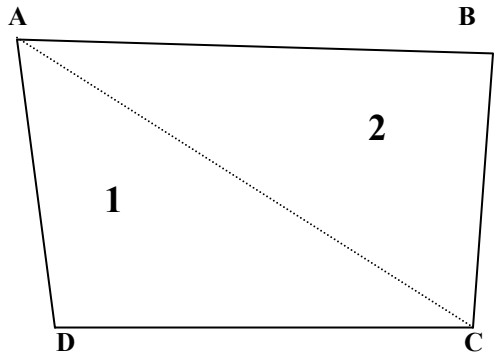


$$s_1 = \frac{(a_1 + b + c_1)}{2}$$

$$s_2 = \frac{(a_2 + b + c_2)}{2}$$

For example, a net with the following dimensions:

Where:
A to B = 81cm
B to C = 73cm
C to D = 71cm
D to A = 76cm
A to C (diagonal) = 109cm



s for Triangle 1: $s = \frac{(71 + 76 + 109)}{2} =$ 131.5

The area of Triangle 1:

$$\sqrt{[131.5 * (131.5 - 81)(131.5 - 73)(131.5 - 109)]} = 2956.5 cm^2$$

s for Triangle 2: $s = \frac{(81 + 73 + 109)}{2} = 128$

The area of Triangle 2:

$$\sqrt{[128 * (128 - 71)(128 - 76)(128 - 109)]} = 2684.9 cm^2$$

The total area of the net would be: $2956.5 cm^2 + 2684.9 cm^2 = 5641.4 cm^2$ or $0.56 m^2$

Figure 7-6. Example of the calculation required to estimate the surface area of water sampled for a ditch net.

8 SOP 8: Measuring Ancillary Environmental Variables

8.1 Water temperature

- Water temperature (ºC) is measured at each sampling station at the time of sampling. Water temperature, to the nearest degree C, can be measured using a stick thermometer or temperature probe.
- Temperature should be taken at mid-depth of the water column.

8.2 Salinity

- Water salinity (ppt) is measured at each sampling station at the time of sampling. Salinity is measured, to the nearest part per thousand, using either a refractometer or water quality probe.
- Salinity should be taken at mid-depth of the water column.

8.3 Dissolved Oxygen

Dissolved oxygen is a common water quality variable that is often collected in conjunction with nekton sampling; however, single measurements are often difficult to interpret and a diurnal time series provides more useful information (Raposa and Roman 2001a). However, since this variable is easily taken with the use of a water probe, we suggest including dissolved oxygen as an ancillary variable.

- Dissolved oxygen in the water (mgl^{-1}) is measured at each sampling station at the time of sampling. Dissolved oxygen, measured in milligrams per liter, can be measured using a water quality probe.
- Dissolved oxygen should be taken at mid-depth of the water column.
- The sample should be taken from an area with little sediment disturbance. It may be necessary to measure a slight distance from the where the throw trap landed or from where the ditch net was pulled to avoid getting erroneous readings due to sediment disturbance caused by the sampling gear.

8.4 Water Depth

Water depth is a simple measure and is useful for documenting changes in water depth over time.

- Water depth (cm) in the throw trap or ditch net is measured to the nearest cm using a meter stick.
- The sides of the trap can be marked off in centimeters and readings taken directly from the trap.
- The trap is often located on an uneven bottom, and thus, depth should be measured near each corner (at least three measurements should be recorded) of the trap to obtain an average depth value.

8.5 Ditch Depth (ditch net only)

- This measurement is useful in determining the flooding stage of the ditch. Depth of the ditch (cm) where the ditch net should be estimated using a meter stick to the nearest cm.
- This measurement is taken from the marsh surface to the bottom of the ditch.

- Water and creek depth for the ditch net are taken in the ditch after the net is removed from the ditch.

8.6 Percent Vegetative Cover (if present)

If macroalgae, aquatic vegetation (*e.g*., *Ruppia*) or eelgrass are present within the throw trap, cover and species composition should be quantified. These data provide a measure of the complexity of habitat available to the estuarine nekton. Since aquatic vegetation is rarely present in ditches, this measure is not recorded for ditch data.

- Prior to dip netting for nekton, the percent cover of each plant species should be visually estimated according to the following cover class categories (<1% cover, 1-5%, 6-10%, 11-25%, 26-50%, 51-75%, >75%).
- If percent cover cannot be estimated due to poor water clarity, then vegetation should be quantified by a biomass technique after Raposa and Oviatt (2000).
    - Algae are placed in plastic bags, returned to the laboratory, identified to species, and dried at 80^{o}C for dry weight determination (the data are expresses as dry weight m^{-2}).
    - Submerged rooted vegetation is quantified by obtaining three cores (25 cm diameter) from immediately outside of the throw trap area.
    - Vegetation collected is sieved in the field to remove sediment, placed in plastic bags, and returned to the laboratory for identification and dry weight determination

9 SOP 9: Data Management

Revision History Log:

Prev. Version #	*Revision Date*	*Author*	*Changes Made*	*Reason for Change*	*New Version #*
Original SOP	*10/14/04*	*Sue Huse*	*Original SOP*		*#1*

This Standard Operating Procedure (SOP) provides detailed instructions for analyzing Salt Marsh Monitoring nekton data collected by the National Park Service Northeast Coastal and Barrier Network (NCBN).. This SOP describes how to create and report data summaries annually, and how to prepare data optional long-term trends and multivariate analyses by researchers as needed for the nekton monitoring.

9.1 Annual Reporting

9.1.1 Automated Reporting

On an annual basis the following analyses will be conducted for nekton monitoring.. The data analyses will include basic species occurrences and metrics. All data will be summarized by marsh within each park for the sampling year. This list should not be interpreted as restricting the inclusion of additional relevant analyses.

The following metrics are reported for nekton:

- Species Occurrence
- Nekton Density
- Nekton Length
- Nekton Counts
- Ancillary Environmental Data (water temperature, salinity, dissolved oxygen)

The Salt Marsh Monitoring Database includes tools that automate the reporting of nekton data for an annual summary table (Fig 9-1).

- Select *Analysis and Export* from the *Main Menu* of the database.
- Selecting *Summary Reports*.
- Select the summary of interest from the list.
- Click *Preview* to view the report or *Print* to print it.

9.1.2 Export Digital Version of Data Summary

A digital version of the summary data can be exported for direct inclusion in a text document or for use in a spreadsheet or other program (Fig. 9-2).

- Select *Export Data to Excel* from the *Analysis and Export* menu.
- Select the summary table of interest .
- Click *Preview* to view the table or *Export* to save it to an Excel formatted file.

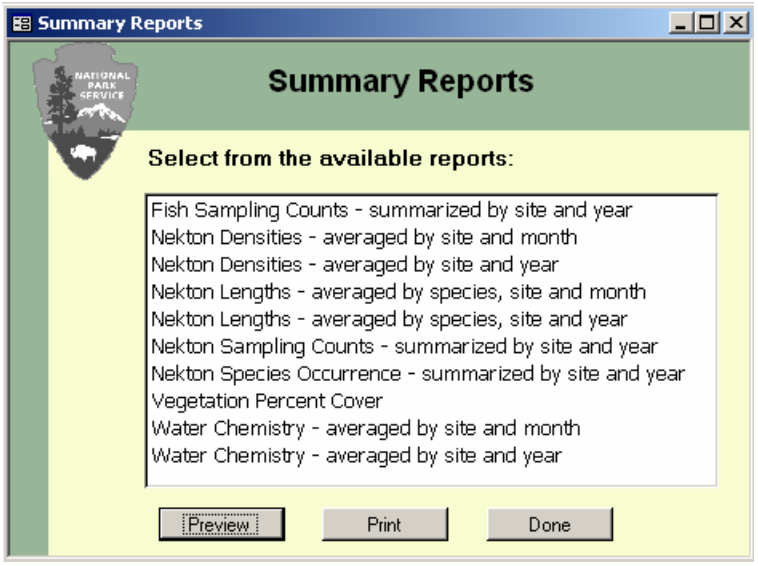


Figure 9-1. Summary reports for printing.

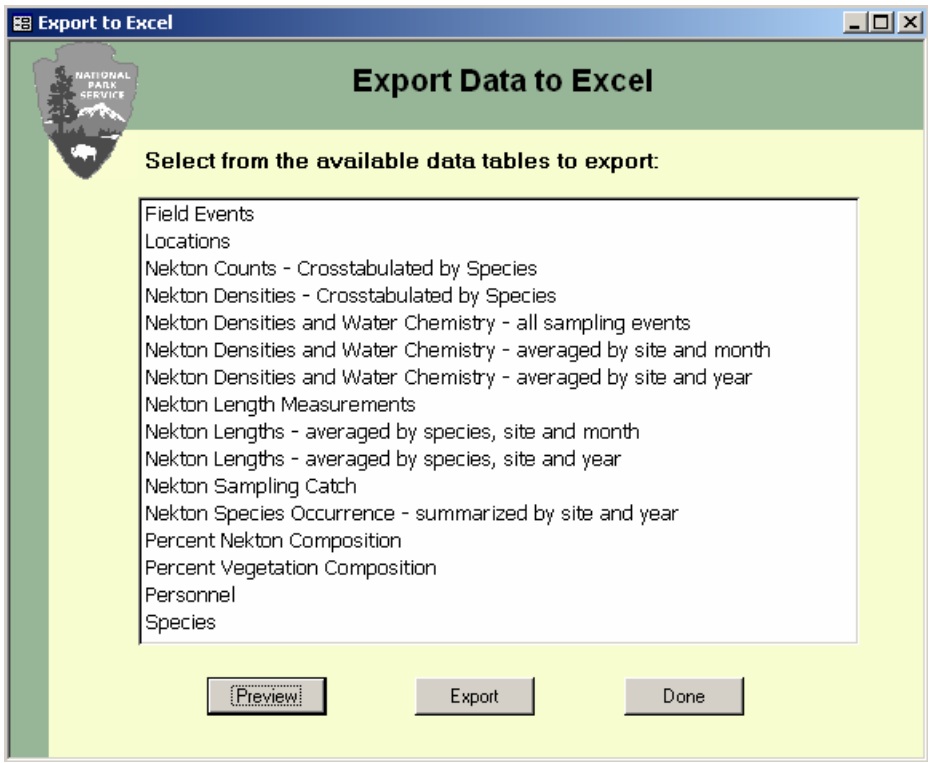


Figure 9-2. Digital export of summary data.

9.2 Multiyear Change Analysis and Comparisons Across Sites

The Salt Marsh Monitoring protocol collects data that can be used to analyze the changes in salt marsh ecology over time, and between monitoring sites and across parks. The protocol includes monitoring each site every three years. The time lag between site visits precludes annual change analyses. Instead, the Principal Investigator and the Network Coordinator will determine how often change analyses should be conducted. The Principal Investigator and the Network Coordinator will also work with park staff to determine if other analyses are required, for which sites, and how often they should be performed. Instructions are included in the sections below for some intermittent analyses.

9.3 Annual Analyses of Nekton Monitoring Data

Analysis of nekton monitoring data include both annual summaries and multi-year analyses. The annual summaries include, but are not limited to species occurrence, nekton density, nekton length, and sampling counts. Multi-year analyses will be conducted every 5 years and will incorporate environmental variables that have been concurrently collected. The Salt Marsh Monitoring Database will not be used to analyze multi-year data. These data will be exported to the applicable statistical program for complete analysis.

9.4 Calculating and Reporting Species Occurrence

Species Occurrence is a list of all nekton species found at each sampling site. The list aggregates the species found at each station by the marsh site where each station is found. The Salt Marsh Monitoring Database includes an automated routine for generating the complete list each year.

9.4.1 Automated Report of Species Occurrence

- From the *Main Menu*, select *Analysis and Export*.
- Then select *Summary Reports*.
- From the list of available reports, highlight *Nekton Species Occurrence – summarized by site and year*.
- Click *Preview* to view the report or *Print* to print it.

9.4.2 Digital Export of Species Occurrence

- Select *Analysis and Export* from the *Main Menu*.
- Select *Export Data to Excel*.
- From the list of available reports, highlight *Nekton Species Occurrence – summarized by site and year*.
- Click *Preview* to view the table or *Export* to save it to an Excel formatted file.

To create a report or export the data for only a subset of the data, you will need to edit the criteria in the base query: "qry_Analysis_SM_NektonSpecies". Follow the instructions in the section "Subsetting Query Data" (Section 8-13). *Be sure to go back to the base query and remove your changes as soon as you have run the report or export!*

9.5 Calculating and Reporting Nekton Density

9.5.1 Calculating Nekton, Fish and Decapod Densities

Creating
Nekton density (individuals per m^2) equals the total number of nekton collected at a station (fish and crustaceans together) divided by area sampled. The sample area is the size of the net or trap used to collect the nekton. Fish and crustacean densities can be calculated individually in the same way.

- The surface area sampled by a throw trap is always $1m^2$.
- The surface area sampled by a ditch net is calculated as the sum of the area of 2 irregular triangles (refer to Section 7.3.2 and Fig. 7-6).

```
Nekton Density by Station =
   (# individuals collected at a station) /
   (surface area sampled (in m2))

Average Nekton Density by Site =
   Σ (Nekton Density per station) by site /
   (Number of stations at the site)
```

Density is calculated for each station individually. If a species is not found at a station, its density equals zero. The densities calculated at each station are averaged together (including the zero densities) to obtain a site average. Density is calculated for each species separately. An estimate of error (standard deviation) and sample size (number of stations sampled) should also be presented. Densities can be presented in a table or graphically. Fish and decapod densities are calculated in the exact same manner.

9.5.2 Automated Nekton Density Reports

The Salt Marsh Monitoring database includes an automated routine for generating the nekton density summaries. The analysis output is available as either a report or an export table format.

- Select *Analysis and Export* from the *Main Menu*.
- Select *Summary Reports* (see Figure 8-1).
- From the list of available reports, highlight *Nekton Densities – averaged by site and year*.
- Click *Preview* to view the report or *Print* to print it.

9.5.3 Export Digital Version of Nekton Densities

- Select *Analysis and Export* from the *Main Menu*.

- Select *Export Data to Excel* (see Figure 8-2).
- From the list of available reports, highlight one of several *Nekton Densities* options.
- Click *Preview* to view the table or *Export* to save it to an Excel formatted file.

9.5.4 To Report or Export a Subset of the Data

To create a report or export the data for only a subset of the data, you will need to edit the criteria in one of the base queries.

- Use "rpt_Analysis_SM_NektonAverages_SiteMonth" and "rpt_Analysis_SM_NektonAverages_SiteYear" for the analyses by site and year or month.
- For export of "Nekton Densities - by Species" use"qry_Analysis_SM_NektonCollections_DensityCrosstab".
- For "Nekton Densities and Water Chemistry - all sampling events" use "qry_Analysis_SM_NektonDensities_WaterChemistry".

Follow the instructions in the section "Subsetting Query Data" (Section 8-13). *Be sure to go back to the base query and remove your changes as soon as you have run the report or export!*

9.6 Long-term Change and Site Comparisons of Nekton Density

An Analysis of Variance (ANOVA) can be used to determine if nekton densities (or fish and crustacean densities) are changing over time or are different among sites (*e.g.,* marshes).

- Usually density data are log transformed [*e.g.*, log (X+1)] prior to statistical analyses to conform to the assumption of normality.
- The dependent variable is density and the independent variable is either year or site, depending on the hypothesis.
- If more than two years or sites are compared then a multiple comparisons post hoc test (*e.g.*, Least Square Means, Tukey) should be used to determine where significant differences are found.

All data should be checked to ensure that the assumptions of the ANOVA are met (*e.g.*, normality, homogeneity of variances). If data do not meet the assumptions of ANOVA then transformations can be conducted or a non-parametric equivalent (*e.g.*, Kruskal-Wallis) can be employed.

9.7 Calculating and Reporting Nekton Species Length

9.7.1 Calculating Nekton Lengths

Average Species Length by Station =

Σ (Lengths of species *i* for each station) /

(Number of species *i* measured at each station)

Average Species Length by Site =

Σ (Species *i* Lengths by station /

(Number of stations where individuals of species *i* were measured at the site)

- Average length of individual species is calculated from the length data for each sampling station.
- All species lengths for each station in the sampling site are included in the average.
- An estimate of error (standard deviation) and sample size (number of individuals measured) should be presented.
- A subsequent analysis is the average length of individual species by site.
- The average nekton length by site is calculated in the same way as the station average, and should also include standard deviation and sample size.
- Lengths may be averaged for all samples of a given species by month or by year. The monitoring protocol includes two field surveys, one in early summer (June) and one later (August).
- For most purposes it is better to report the average lengths separately for each month, although both reports are available.

9.7.2 Automated Nekton Length Reports

The Salt Marsh Monitoring database includes an automated routine for generating the nekton length summaries. The analysis output is available as either a report or an export table format.

- Select *Analysis and Export* from the *Main Menu*.
- Select *Summary Reports*.
- From the list of available reports, highlight *Nekton Lengths- averaged by species, site and month*, or *Nekton Lengths- averaged by species, site and year*.
- Click *Preview* to view the report or *Print* to print it.

9.7.3 Export a Digital Version of Nekton Lengths

A digital version of these data can be exported for direct inclusion in a text document or for use in a spreadsheet or other program.

- Select *Analysis and Export* from the *Main Menu*.
- Select *Export Data to Excel*.
- From the list of available reports, highlight *Nekton Lengths- averaged by species, site and month*, or *Nekton Lengths- averaged by species, site and year*.
- Click *Preview* to view the table or *Export* to save it to an Excel formatted file.

9.7.4 To Report or Export a Subset of the Data

To create a report or export the data for only a subset of the data, you will need to edit the criteria in one of the base queries.

- Use "rpt_Analysis_SM_NektonLengths_SiteMonth" and "rpt_Analysis_SM_NektonLengths_SiteYear" for the analyses by site and year or month.
- For export of "Nekton Lengths - all data by species" use "qry_Analysis_SM_NektonLengths_Crosstab".

Follow the instructions in the section "Subsetting Query Data" (Section 8-13). *Be sure to go back to the base query and remove your changes as soon as you have run the report or export!*

9.8 Multiyear Change Analysis of Nekton Lengths

Distribution analyses (*e.g.*, Kolmogorov-Smirnov Test) will be employed to determine if length-frequency distributions of a species are changing over time.

If multiple comparisons among size-frequency distributions are made for the same species then alpha levels should be adjusted using a Bonferroni correction (Zar 1999) or step-wise Bonferroni correction (Rice 1989). For example, the Bonferroni correction for 4 pair-wise comparisons at a probability level is 0.05, would result in an adjusted alpha level of 0.05/4 or 0.0125. Any comparisons having p-values below 0.0125 would be significantly different.

9.9 Calculating and Reporting Individual Species Sampling Counts

Counts of the number of individuals per species are also calculated for inclusion in reports

9.9.1 Calculating Sampling Counts

Species Count by Site =

Σ (Number of individuals of species *i* collected per station) by site

9.9.2 Automated Annual Individual Species Count Reports

To create a printable report, select *Analysis and Export* from the *Main Menu*, then select *Summary Reports*. From the list of available reports, highlight *Fish Sampling Counts - summarized by site and year*. Click *Preview* to view the report or *Print* to print it.

To export a digital version of this data for direct inclusion in a text document or for use in a spreadsheet or other program, select *Analysis and Export* from the *Main Menu*, then select *Export Data to Excel*. The fish count data are included in any of the *Nekton Densities and Water Chemistry* exports. The fish count data are included in the query with the other nekton density data. Click *Preview* to view the table or *Export* to save it to an Excel formatted file.

To create a report or export the data for only a subset of the data, you will need to edit the criteria in one of the base queries. Use "qry_Analysis_SM_NektonAverages_SiteMonth" and "qry_Analysis_SM_NektonAverages_SiteYear" for the analyses by site and month or year.

Follow the instructions in the section "Subsetting Query Data" (Section 8-13). . *Be sure to go back to the base query and remove your changes as soon as you have run the report or export!*

9.10 Export of Nekton and Environmental Variables

For this export, the table fields (column headings) are: park, site, year, date, station, gear type (throw trap or ditch net), habitat, nekton density, fish density, crustacean density, water temperature, salinity, dissolved oxygen, and water depth.

This table includes a record (row) for every sampling event – by location and date. If a sampling event yields a count of zero, no nekton present, the event is to be included in the data with a value of 0.

- To export this table, select *Analysis and Export* from the *Main Menu*.
- Select *Export Data to Excel*.
- From the list of available reports, highlight *Nekton Densities and Water Chemistry - all sampling events*.
- Click *Preview* to view the table or *Export* to save it to an Excel formatted file.

To create a report or export the data for only a subset of the data, you will need to edit the criteria in the base query: "qry_Analysis_SM_NektonDensities_WaterChemistry".

Follow the instructions in the section "Subsetting Query Data". *Be sure to go back to the base query and remove your changes as soon as you have run the report or export!*

9.10.1 Export Individual Nekton Densities

Community analyses require information on the individual species densities for each sampling event. The previous table combined densities for nekton, fish and decapods across species. This table will contain the fields (column headings) Park, Site, Date, Year, Gear, Station, Habitat and then a column for each species found. Each table record (row) represents a sampling event. The data are the species densities for each species at each station. This format is most flexible for use with analytical software such as PRIMER.

- To export this table, select *Analysis and Export* from the *Main Menu*.
- Select *Export Data to Excel*.
- From the list of available reports, highlight *Nekton Densities by Species*.
- Click *Preview* to view the table or *Export* to save it to an Excel formatted file.

> To export the data for only a subset of the data, you will need to edit the criteria in the base query: "qry_Analysis_SM_NektonCollections_DensityCrosstab".
>
> Follow the instructions in the section "Subsetting Query Data" (Section 8-13). . *Be sure to go back to the base query and remove your changes as soon as you have run the report or export!*

9.10.2 Export Individual Nekton Lengths

Individual nekton length data can show the growth of individual nekton species and how this may vary by site and year. The structure of this table is similar to the individual nekton densities export table above. The table contains the fields (column headings): Park, Site, Date, Year, Gear, Station, Habitat and then a column for each species whose length was measured. Each table record (row) represents one measured nekton for one sampling event. The data are the length of the each species at each station. An estimate of error (standard deviation) and sample size (number of stations sampled) should also be presented.

- To export this table, select *Analysis and Export* from the *Main Menu*.
- Select *Export Data to Excel*.
- From the list of available reports, highlight *Nekton Lengths - all data by species*.
- Click *Preview* to view the table or *Export* to save it to an Excel formatted file.

To export the data for only a subset of the data, you will need to edit the criteria in the base query: "qry_Analysis_SM_NektonLengths_Crosstab".

Follow the instructions in the section "Subsetting Query Data" (Section 8-13). . *Be sure to go back to the base query and remove your changes as soon as you have run the report or export!*

9.11 Environmental Data

9.11.1 Annual Summary of Environmental Data Averages

```
Environmental Variable Averages =
   (Σ Variable values per station at a site) /
   (Number of stations at the site)
```

The average value for the measured environmental variables (water temperature, salinity, dissolved oxygen) equals the sum of the value at each sampling station in a site divided by the number of stations in that site. An estimate of error (standard error or standard deviation) and sample size (number of stations sampled) should be presented. Averages are calculated separately for each month or year.

9.11.2 Automated Report for Environmental Variables

The Salt Marsh Monitoring database includes an automated routine for generating the environmental data averages. The analysis output is available as either a report or an export table format.

- Select *Analysis and Export* from the *Main Menu*.
- Select *Summary Reports*.
- From the list of available reports, highlight *Water Chemistry - averaged by site and year*, *Water Chemistry – averaged by site and month*, or *Nekton Densities and Water Chemistry - all sampling events*.
- Click *Preview* to view the report or *Print* to print it.

9.11.2.1 Export a Digital Version of Data

- Select *Analysis and Export* from the *Main Menu*.
- Select *Export Data to Excel*.
- From the list of available reports, highlight the table you would like to export.
- Click *Preview* to view the table or *Export* to save it to an Excel formatted file.

9.11.3 Report or Export a Subset of the Data

To create a report or export the data for only a subset of the data, you will need to edit the criteria in one of the base queries.

- Use “rpt_Analysis_SM_NektonAverages_SiteMonth” and “rpt_Analysis_SM_NektonAverages_SiteYear” for the analyses by site and year or month.
- For “Nekton Densities and Water Chemistry - all sampling events” use “qry_Analysis_SM_NektonDensities_WaterChemistry”.

Follow the instructions in the section "Subsetting Query Data" (Section 8-13). *Be sure to go back to the base query and remove your changes as soon as you have run the report or export!*

9.12 Multiyear Change Analysis and Site Comparisons of Environmental Data

An Analysis of Variance (ANOVA) can be used to determine if environmental variables of sampling sites (water temperature, salinity, dissolved oxygen) are changing over time or are different among sites (*e.g.,* marshes). The dependent variable would be density and the independent variable would be either year or site, depending on the hypothesis. If more than two years or sites are compared then a post hoc test (*e.g.*, Least Square Means, Tukey) should be used to determine where significant differences are found.

All data should be checked to ensure that the assumptions of the ANOVA are met (*e.g.*, normality, homogeneity of variances). If data do not meet the assumptions of ANOVA then transformations can be conducted or a non-parametric equivalent (*e.g.*, Kruskal-Wallis) can be employed.

9.12.1 Export of Multiyear Data

- Select *Analysis and Export* from the *Main Menu*.
- Select *Export Data to Excel*.
- From the list of available reports, highlight *Nekton Densities and Environmental Data – all sampling events*.
- Click *Preview* to view the table or *Export* to save it to an Excel formatted file.

9.13 Subsetting Query Data

The Salt Marsh Monitoring database includes a large number of analytical queries and reports for annual reporting and importing to other analytical software packages. There will be times, when researchers and park staff may want the data but for only a subset of the entire regional project. Obvious examples of this will be exporting data for only the current year, displaying data for one park, or for more specific analysis summarizing specific locations within one site. To subset the data, the user will need to edit the criteria in the appropriate query before exporting or printing the selected output.

9.13.1 Backing up the database front end interface

If you haven't done so already, it is a good idea to backup the database front-end before editing any queries. This cannot be done from within the database. The backup options available on startup and from the main menu are only for the backend data file.

To backup the front end, make a copy of the *MonitoringSM.mdb* file.

9.13.2 Opening the Query and Determine the Query Name

For each of the reporting options described throughout this SOP, queries are used to compile and analyze the data.

- To determine the name of the query you need to edit, review the relevant section of this SOP, where the name of the base query will be listed.
- Query names will usually start with "qry_Analysis_SM*".

9.13.3 Open the database window

The database window displays the list of tables, queries, reports, etc. This window is usually hidden in the Salt Marsh Monitoring database to avoid confusion.

- To open the database window, select*Unhide* from the *Window* menu at the top of the Access application.
- Select the MonitoringSM database.
- Click *OK*.

9.13.4 Open the Query in Design view

- From the list of objects along the left side of the database window, select *Queries*.
- The right side of the window will display the list of all available queries.
- Highlight the query you need to edit.
- With the query highlighted, click the design view button in the upper left of the window.

9.13.5 Editing the Query

The design view of a query will show you the queries and tables whose data are the input to the query, and how each of fields is defined.

- If you click the view button in the far left of the toolbar, you can see the query output in datasheet view or return to the design view.
- The design view has two main sections. The upper section shows the tables or queries that are input and how they relate to one another.
- The lower section defines the output fields and criteria. *Only edit the lower section criteria in the design view*.
- All further directions below refer to the lower section only.

9.13.6 Check for existing criteria

This step is critically important! Before you begin editing criteria, you must check to see what criteria are already included in the query. For instance a nekton vs vegetation may include *protocol = "SMN"*. *Any field that already has a criteria, you should not edit!* If you edit existing criteria, the dependent queries and reports will no longer be valid. Be sure you know which criteria are part of the original query, and *do not remove these when you reset the query!*

9.13.7 Determine the fields to subset

Along the left of the window are the row identifiers: *Field, Table, Sort*, etc. The top row is the field row and this includes the field names and definitions. A colon is used to separate a field name from its definition. If there is no colon, the field name is whatever string is listed in that cell. From the list of fields, determine which you need to edit. In this example, the field names are: *Park, Site, Station, Year, and Method*. To include only

data from 2004, you will need to edit the year field. To restrict the data to "King Creek" in "Colonial National Park", you will need to edit both the *Park* and *Site* fields.

9.13.8 Determine the field values to express

To write out specific criteria, you need to know the field values. In the above example, if you want to include only data from Colonial National Park, you need to know if the query values for Colonial National Park are "Colonial", "Colonial National Park", or "COLO".

If you are unsure of the exact format of the values you need, return to the *Datasheet view* by clicking on the view button as described above. Scroll through the data until you see the values you are looking for. Then return to the *Design view*, and continue. In the figure below, you can see that the park value for Colonial is "COLO"

9.13.9 Enter the criteria

The type of criteria you are using determines how it will be expressed. In all cases, the criteria will be entered into the *Criteria* row.

9.13.9.1 Entering exact values

An exact value will be where you know what the value of the field data are exactly. There may be more than one value, but you can express the value in exact terms. In the example above, the park value is "COLO". Year would be 2003.

Once you know the exact value you want you need to enter it into the Criteria row. Enter text values with quotations and numeric values without.

To enter more than one value for a given field, say Colonial, Boston Harbor Islands, and Fire Island, use the *Or* and subsequent rows under *Criteria*.

9.13.9.2 Entering a range of text values

An example where this is useful is in subsetting stations within a site. This works using wildcard values, when using the *Or* is unrealistic. For example, in 2004, three transects were used for measuring vegetation data with the 50 point intercept method. The first transect has 13 stations, the second has 10 stations, and the third has 9. To include only data from transect 1 would require 13 *Or* statements or one wildcard statement.

The BOHA stations names are the year, the transect and the distance along the transect. So, a distance of 10 meters along transect 1 in 2004, is station "04_T1-10". To include all T1 stations, use a wildcard expression such as "*T1*" To be sure that you only include BOHA stations, enter criteria for park and site as well.

When entering wildcard expressions as criteria, it is necessary to include the word *Like* before the expression so Access will interpret it as an approximation with wildcards rather than an exact value.

9.13.9.3 Entering numeric ranges

Set up numeric range criteria just as you would in standard math notation. For instance, to select all percent cover measurements between 50 and 75% enter >=50 And <=75. Remember quotations are for text values only.

9.13.9.4 Entering dates

Set up your date criteria just as you would the other criteria, but you will need to bracket dates with #'s just as you would use quotes to bracket text. For example to include only data from June 2004, your criteria would be >=#6/1/2004# And <=#6/30/2004#.

9.13.10 Check your criteria

To see if you have entered your criteria correctly, switch to *Datasheet view* and scroll through your data. If you have an empty query, you have entered an invalid criteria for which no data have that value. This can easily be caused by a misspelling. Or perhaps the answer is there are no data meeting your criteria. If you do not see the data you expect, recheck your criteria. You may need to remove all your criteria and review the original query to determine if you are having difficulty with the data or with your criteria.

9.13.11 Save and close

Once you have entered your criteria, you must save the query. Click the save button in the upper left corner of the Access application window. Close the query

9.14 View the output

9.14.1 Return to the output menus.

Bring the *Main Menu* forward again, and select *Analysis and Export*. Select either *Summary Reports* or *Export Data to Excel*, depending on which data you are interested in.

9.14.2 Preview the new data

Select the export or report data and click *Preview*. If the data output is as you export, you are ready to print or export. If the data are not as you expect, review your criteria-setting steps above. If the data are still not what you expect, contact your Data Administrator for further assistance.

9.15 Remove the query criteria

When you have finished and printed or exported the subsetted data you need, *be sure to return the query to its original form!* If you do not remove your subsetting criteria, other users, or yourself will have unexpected results when using the data export and reporting tools. This may be weeks or even a year later, long after these steps have been forgotten. It is particularly important to do it sooner, rather than later, because some of the criteria in the query may be part of the original, and should not be removed. If you do not clean up your work immediately, other users, or even yourself, will have no way to know which criteria should be kept and which removed.

Using the directions above as needed, open the query in design view again. Delete each of the criteria you have entered. Save and close the query.

9.16 Quality Control in Annual Data Reports

The series of automated annual reporting summaries, have undergone a quality control review during development. When using these annual reporting tools, it is imperative that researchers continue to review these data summaries each time they are used.

9.16.1 Development Quality Control

The Network has performed quality control on the summary reports prior to their release. This quality control consists of cross-checking the following items:

- Field names and values – are the necessary fields included in the summaries, and do these fields display the appropriate information? Each field name and the values reported is checked for all queries and reports.

- Record counts – do the summary queries and reports have the correct number of records? The number of records in each output query is compared to the number of records in the input tables and queries. Insufficient record counts may stll arise if not all of the field data has been entered into the database.

- Sample counts –does each average or other summary calculation have the correct sample number? Summary statistics combine data from a series of events, usually by site and year. The number of events combined for that statistic for that site and year is the sample number. Sample numbers are spot-checked.

- Sample sums – do the reported totals equal the sum of the data values? Totals are spot-checked for various subsets of the data, based on the summary.

- Summary values- are the summary statistics accurate?

Summary statistic values are also spot-checked. If independent calculations are available, the summary values are compared with the independent values. Where independent summary values are not available, spot checks are made. Particularly with averages, since most queries will include both the total and the sample count, both of which have been checked.

9.17 Reporting Quality Control

Each time the summary data are exported for inclusion in an annual report, the individual responsible for reporting must perform a basic quality control check before disseminating the report data. Even though the analysis development has been checked, it is important for the specific data report values to be checked as well. This will help detect errors in data entry and any changes made to the summaries through subsetting of the base queries.

- Data entry quality control – before running summary analyses and checking them for accuracy, it is necessary to perform quality control on the data entry. If the data have been entered with inaccurate data, or if data entries are missing, the summary analyses will be incorrect.

- Data aggregation units – are all the parks, sites, locations and dates that you are reporting on included in the summary? If not, be sure to check the base query to be sure that no subsetting remains from a previous report.

- Record counts – depending on the type of summary or export, you cross-check against the number of field events. Otherwise, do you have data reported for each park and site? If data are not summarized by site, do you have data for each sampling location? If locations were visited more than once during the year, do you have matching data from each sampling trip?

- Sample counts – if you are summarizing by site, do you have the correct number of locations included in your sample count? If you are reporting averages, do you have the correct number of sample counts for each event. If you have more than one data value for an event, (e.g., nekton sampling lengths), do you have the correct number of samples per location (e.g., compare sample n for nekton lengths, with the sample n for nekton collection).

- Sample sums – spot check the totals by performing the calculation independently for a few of the data values.

- Summary values – spot check the values by performing the calculation independently for a few of the data values. With averages this can be particularly easy if both the total and the sample counts are also reported.

10 SOP 9: Data Analyses

10.1 Nekton Density Data

10.1.1 Annual Analyses

- Species lists should be made for each sampling site (*i.e.*, marsh).
- Nekton density (number of individuals per m^2) is calculated as the total number of nekton (fish and crustaceans can either be analyzed separately or together) divided by area sampled (*i.e.*, $1m^2$ for throw traps or the calculated area for ditch nets).
- Density is calculated for each station, and all stations (stations with no nekton collected are included as zeros) are averaged together to obtain a site average. Density for each individual species can similarly be calculated.
- An estimate of error (standard error or standard deviation) and sample size (number of stations sampled) should be presented.

10.1.2 Trend Analyses

- An Analysis of Variance (AVOVA) can be used to determine if nekton densities (or fish and crustacean densities) are changing over time or are different among sites (*e.g.,* marshes). Usually density data are log transformed [*e.g.*, log (X+1)] prior to statistical analyses to conform to the assumption of normality. The dependent variable would be density and the independent variable would be either year or site, depending on the hypothesis. If more than two years or sites are compared then a post hoc test (*e.g.*, Least Square Means, Tukey) should be used to determine where significant differences are found.
- All data should be checked to ensure that the assumptions of the ANOVA are met (*e.g.*, normality, homogeneity of variances).
- If data do not meet the assumptions of ANOVA then transformations can be conducted or a non-parametric equivalent (*e.g.*, Kruskal-Wallis) can be employed.

10.2 Nekton Community Data

10.2.1 Annual Analyses

- Species lists should be made for each sampling site (*i.e.*, marsh).

10.2.2 Trend Analyses

- Additional analyses that we often use are part of the PRIMER software package (http://www.primer-e.com), that use non-parametric tests to detect differences in community structure (*i.e.*, species composition and abundance). Non-parametric permutation testing procedures can be effectively used to evaluate dissimilarity or similarity in nekton communities between marshes or between sample years. ANOSIM, part of the PRIMER statistical package (Plymouth Routines In Multivariate Research, Carr 1997) is just one example of a non-parametric test, similar to multivariate analysis of variance (MANOVA) but without the generally unattainable assumptions (Clarke and Warwick 1994, Carr 1997). The ANOSIM

procedure calculates a similarity measure (such as the Euclidean Distance measure), and a similarity matrix is created that allows for the objective identification of samples (*e.g.*, nekton sampling stations) that have similar (or dissimilar) communities in terms of species composition and abundance. All pair-wise comparisons are summarized into a test statistic using Clark's R that compares between-group to within-group dissimilarities. Monte Carlo permutation tests are then used to derive p-values.

- Pairwise comparisons between groups of samples are defined *a priori* to detect differences in communities (*e.g.*, 2001 vs. 2002).
- A Bonferroni correction (Zar 1999) or step-wise Bonferroni correction (Rice 1989) for the experiment-wise error is made based on the number of comparisons being tested. For example, the Bonferroni correction for 4 pair-wise comparisons at a probability level is 0.05, would result in an adjusted alpha level of 0.05/4 or 0.0125. Any comparisons having p-values below 0.0125 would be significantly different.

- For nekton community composition analyses we use the defaults of the program (no standardization, no transformation), and the Euclidean distance metric.
- For pairwise comparisons that are significant, or have dissimilar communities, it is often desirable to know what contribution the individual cover types or species made to the dissimilarity. The proportion of the overall dissimilarity that is contributed by individual species can be calculated as follows;

Where;

$$1-\frac{D}{D\max}=1-\frac{(C_{1i}-C_{2}i)^{2}}{\sum(C_{1i}-C_{2i})^{2}}$$

D = Distance
C_{1i} = abundance of species i in marsh 1
C_{2i} = abundance of species i in marsh 2

- The outcome is a list of species ranked in order of their percent contribution to the dissimilarity between significant pairwise comparisons. D_{max} (based on Euclidean Distance) provides an overall measure of dissimilarity for each pairwise comparison. D_{max} values can be used to determine if communities on different marshes are becoming more similar. For example, as D_{max} values become more alike (*i.e.*, closer together), this is indicates that the communities of the marshes are becoming more similar. Conversely, as D_{max} become farther apart, this indicates that communities are becoming more dissimilar.

10.3 Nekton Length Data

10.3.1 Annual Analyses

- Average length of individual species for a study site is calculated from the length data for each sampling site.

- An estimate of error (standard error or standard deviation) and sample size (number of individuals measured) should be presented.

10.3.2 Trend Analyses

- Distribution analyses (*e.g.*, Kolmogorov-Smirnov Test) can be employed to determine if length-frequency distributions of a species are changing over time.
- If multiple comparisons among size-frequency distributions are made for the same species then alpha levels should be adjusted using a Bonferroni correction (Zar 1999) or step-wise Bonferroni correction (Rice 1989). For example, the Bonferroni correction for 4 pair-wise comparisons at a probability level is 0.05, would result in an adjusted alpha level of 0.05/4 or 0.0125. Any comparisons having p-values below 0.0125 would be significantly different.

10.4 Environmental Data

10.4.1 Annual Analyses

- An average for each of the environmental variables (water temperature, salinity, dissolved oxygen) is calculated for the study site.
- An estimate of error (standard error or standard deviation) and sample size (number of stations sampled) should be presented.

10.4.2 Trend Analyses

- An Analysis of Variance (AVOVA) can be used to determine if environmental variables of sampling sites (water temperature, salinity, dissolved oxygen) are changing over time or are different among sites (*e.g.,* marshes). The dependent variable would be density and the independent variable would be either year or site, depending on the hypothesis. If more than two years or sites are compared then a post hoc test (*e.g.*, Least Square Means, Tukey) should be used to determine where significant differences are found.
- All data should be checked to ensure that the assumptions of the ANOVA are met (*e.g.*, normality, homogeneity of variances).
- If data do not meet the assumptions of ANOVA then transformations can be conducted or a non-parametric equivalent (*e.g.*, Kruskal-Wallis) can be employed.

11 SOP 11: Reporting and Review (placeholder for Network)

12 SOP 12: Completion of Field Season: Procedures for Equipment Storage

12.1 Maintenance and Repairs

- All sampling equipment should be cleaned and repaired (if required) prior to storage. Proper storage will help maintain the life of equipment for future sampling endeavors.
    - Ditch nets should be stored with the net wrapped around the oak stakes with the stakes sticking out (*i.e.*, the net should be clear of the stakes to prevent tears in the netting) and should be stored in an area that is free of rodents (mice like to nest in the netting).
- Batteries should be removed from all electronic equipment when not in use for extended periods of time.

13 SOP 13: Revising the Protocol or SOP (placeholder for Network)

This protocol is a revision of a protocol first developed by Raposa and Roman (2001a) for use in the Long-term Coastal Monitoring Program at Cape Cod National Seashore. The original protocol can be found at the National Park Service Inventory and Monitoring website: http://www.nature.nps.gov/im/monitor/protocoldb.cfm

This protocol was revised for the following reasons:

- To conform to NPS Inventory and Monitoring Program format guidelines
- To include the additional sampling method of the ditch net sampler

This protocol was revised September 2004 by:

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Previous Version	Revision Date	Author	Changes Made	Reason for Change	New Version #
Original Protocol	12/13/04	Mary-Jane James-Pirri mjjp@gso.uri.edu	Format Changes; Addition of ditch net SOP	Conform to NPS guidelines; Add ditch net as gear type	#1

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Appendix

This appendix present the UTM (Nad 83, meters) of sampling (nekton and vegetation) sites for each park monitored to date.

Table 1. Coordinates for locations sampled at BOHA in 2004, UTM, Zone 18, NAD 83, meters. ** Indicates that original GPS coordinates were not in correct location and new coordinates were estimated from GIS.*

Site	Sampling variable	Station	UTM X (east)	UTM Y (north)
Thompson Island	Nekton	1	333828	4686459
		2	333885	4686484
		3	333904	4686479
		4	333934	4686468
		6	333946	4686447
		7	333971	4686437
		8	333975	4686463
		9	333990	4686482
		10	333981	4686509
		11	334018	4686441
		12	334039	4686480
		13	334078	4686488
		14	334085	4686310
		15	334100	4686361
		16	334119	4686393
	Vegetation	1-00	333869	4686512
		1-10	333863	4686496
		1-20	333864	4686482
		1-30	In creek no data recorded	
		1-40	333868	4686463
		1-50	333869	4686452
		1-60*	333870	4686442
		1-70*	333871	4686422
		1-80	333870	4686397
		1-90	In creek no data recorded	
		1-100	In creek no data recorded	
		1-110	In creek no data recorded	
		1-120	333875	4686350
		1-130*	333876	4686338
		2-00	333915	4686510
		2-10	333915	4686488
		2-20	333921	4686483
		2-30	In creek no data recorded	
		2-40	333924	4686465

Site	Sampling variable	Station	UTM X (east)	UTM Y (north)
		2-50	333929	4686451
		2-60*	333930	4686441
		2-70	333932	4686431
		2-80*	333933	4686424
		2-90	333936	4686418
		2-100	333957	4686294
		3-00	334000	4686541
		3-10	In creek no data recorded	
		3-20	In creek no data recorded	
		3-30	334006	4686509
		3-40	334008	4686498
		3-50	334008	4686488
		3-60	In creek no data recorded	
		3-70	334019	4686464
		3-80*	334022	4686453
		3-90	334080	4686313
		3-100	In creek no data recorded	
		3-110	334083	4686286
		3-120*	334085	4686272
Calf Island	Vegetation	1-00*	343785	4689492
		1-10	343788	4689481
		1-20	343793	4689477
		1-40	343796	4689456
		1-50	343800	4689448
		1-60*	343805	4689438
		1-70	343812	4689430
		1-80	343820	4689424
		1-90	343816	4689416
		1-100	343826	4689413
		1-110	343822	4689408
		2-00	343805	4689491
		2-10	343807	4689482
		2-40	343826	4689444
		2-50	343833	4689425
		2-60	343833	4689432
		2-70	343838	4689422
		2-80*	343843	4689414

Table 2. Coordinates for locations sampled at CACO in 2004, UTM, Zone 18, NAD 83, meters.

Site	Habitat	Station	UTM X (east)	UTM Y (north)
Nauset Marsh	Creek	C1	421320	4629508
		C2	421334	4629823
		C3	421338	4629921
		C5	421162	4629202
		C6	421171	4629289
		C7	421217	4629713
		C8	421163	4630141
		C9	421064	4629491
		C10	421032	4629587
		C11	420914	4630300
		C12	420875	4630391
		C13	420963	4629583
		C14	420975	4629801
		C15	420955	4630140
		C17	420885	4629948
		C18	420886	4630083
		C19	420884	4630492
		C21	420726	4630122
	Pool	P1	421337	4629434
		P2	421360	4629632
		P3	421319	4629952
		P4	431309	4629990
		P5	421206	4629353
		P6	421222	4629373
		P7	421251	4629721
		P8	421212	4629994
		P9	421031	4629719
		P10	421034	4629807
		P11	420967	4630062
		P12	420947	4630136
		P13	420990	4629616
		P14	420968	4629953
		P15	420936	4630129
		P17	420889	4630290
		P18	420885	4630410
		P19	420896	4630459
		P20	421267	4630138
		P21	420735	4630067

Table 3. Coordinates for locations sampled at COLO in 2003, UTM, Zone 18, NAD 83, meters. *Note: Back River P1 was not sampled because it was too deep; Back River P5 was not sampled in August because it was too shallow; King Creek, P6 and P7 do not exist. * UTM coordinates of Back River, P6 were estimated from GIS maps.*

Marsh	Sampling Variable	Station	UTM X (east)	UTM Y (north)
Back River	Nekton	P2	342860	4120447
		P3	342876	4120475
		P4	342876	4120494
		P5	342769	4120684
		P6*	342755	4120665
		P7	342731	4120643
		P8	342764	4120632
		P9	342685	4120632
		P10	342659	4120637
		P11	342636	4120607
		P12	342565	4120595
	Vegetation	1-00	342938	4120495
		1-30	342938	4120471
		1-60	342950	4120433
		1-90	342940	4120409
		2-00	342910	4120530
		2-30	342902	4120500
		2-60	342901	4120471
		2-90	342898	4120452
		2-120	342896	4120432
		3-00	342799	4120727
		3-50	342792	4120680
		3-100	342784	4120631
		3-150	342775	4120582
		3-200	342769	4120536
		4-00	342736	4120752
		4-50	342731	4120713
		4-100	342724	4120660
		4-150	342717	4120611
		4-200	342711	4120560
King Creek	Nekton	P1	357640	4126486
		P2	357725	4126459
		P3	357748	4126479

Marsh	Sampling Variable	Station	UTM X (east)	UTM Y (north)
King Creek (continued)	Nekton (continued)	P4	357764	4126448
		P5	357774	4126425
		P8	357813	4126408
		P9	357800	4126382
		P10	357823	4126370
		P11	357862	4126362
		P12	357858	4126401
		P13	357853	4126422
		P14	357856	4126446
		P15	357835	4126512
		P16	357835	4126488
	Vegetation	1-00	357746	4126364
		1-50	357715	4126402
		1-100	357684	4126446
		2-00	357799	4126349
		2-50	357767	4126386
		2-100	357736	4126427
		2-150	357704	4126462
		3-00	357926	4126339
		3-50	357898	4126368
		3-100	357865	4126411
		3-150	357832	4126453
		3-200	357804	4126489
		3-250	357771	4126526
		4-00	357907	4126439
		4-50	357878	4126482
		4-100	357849	4126520
		4-150	357819	4126564

Table 4. Coordinates for locations sampled at FIIS in 2003, UTM, Zone 18, NAD 83, meters. * *UTM coordinates of nekton stations, Hospital Point, D1 to D5 are unavailable, however they were on the two eastern most ditches of the study site; station P8 was estimated from GIS maps. Note: Hospital Point, P4 and P5 do not exist; Watch Hill, D10, D11, and P1 were not sampled in August due to low water.*

Marsh	Sampling Variable	Station	UTM X (east)	UTM Y (north)
Hospital Point	Nekton	D1	Not recorded	
		D2	Not recorded	
		D3	Not recorded	
		D4	Not recorded	
		D5	Not recorded	
		D6	678020	4510820
		D7	677967	4510920
		D8	677916	4511024
		D9	677958	4510796
		P1	678072	4510965
		P2	678051	4510971
		P3	678050	4510985
		P6	678087	4510997
		P7	678117	4510958
		P8*	677981	4510884
		P9	677947	4510846
		P10	677976	4510832
		P11	677974	4510822
	Vegetation	1-00	677997	4510628
		1-50	677967	4510671
		1-100	677944	4510717
		1-150	677912	4510758
		1-200	677886	4510801
		1-250	677856	4510839
		2-00	678020	4510692
		2-50	677997	4510738
		2-100	677972	4510782
		2-150	677954	4510827
		2-200	677933	4510866
		2-250	677911	4510906
		2-300	677882	4510964

Marsh	Sampling Variable	Station	UTM X (east)	UTM Y (north)
Hospital Point (continued)	Vegetation (continued)	2-350	677862	4511021
		3-00	678145	4510705
		3-50	678125	4510750
		3-100	678100	4510801
		3-150	678081	4510847
		3-200	678058	4510887
		3-250	678039	4510942*
		3-300	678013	4510983
		3-350	677992	4511024
		3-400	677966	4511070
		4-00	678199	4510740
		4-50	678176	4510787
		4-100	678153	4510830
		4-150	678123	4510877
		4-200	678102	4510915
		4-250	678083	4510968
		4-300	678067	4511001
Watch Hill	Nekton	D1	670399	4506789
		D2	670391	4506859
		D3	670457	4506817
		D4	670450	4506888
		D5	670468	4506815
		D6	670514	4506832
		D7	670511	4506872
		D8	670524	4506881
		D9	670528	4506859
		D10	670577	4506868
		D11	670570	4506914
		P1	670593	4506908
		P2	670507	4506950
		P3	670463	4506925
		P4	670443	4506916
		P5	670408	4506894
	Vegetation	1-00	670380	4506756
		1-30	670369	4506793
		1-60	670360	4506824
		1-90	670348	4506844
		2-00	670414	4506762

Marsh	Sampling Variable	Station	UTM X (east)	UTM Y (north)
Watch Hill (continued)	Vegetation (continued)	2-30	670414	4506791
		2-60	670417	4506820
		2-90	670418	4506849
		2-120	670419	4506878
		3-00	670459	4506776
		3-30	670459	4506811
		3-60	670452	4506839
		3-90	670453	4506859
		3-120	670451	4506894
		4-00	670510	4506779
		4-30	670511	4506811
		4-60	670508	4506841
		4-90	670501	4506870
		4-120	670508	4506898
		4-150	670511	4506928

Table 5. Coordinates for locations sampled at GATE in 2003, UTM, Zone 18, NAD 83, meters. *Note: The following stations were not sampled in August because of low water: Big Egg Control: NC-18, NC-22, NC-25; Big Egg Treatment: NT-7, NT-11, NT-14; Horseshoe Cove: P4, P9, P14, P16, P19. * Indicates UTM coordinates of vegetation stations were estimated from GIS maps.*

Marsh	Sampling Variable	Station	UTM X (east)	UTM Y (north)
Big Egg Treatment	Nekton	NT-1	599311.7	4494539
		NT-2	599277.9	4494539
		NT-3	599252.5	4494539
		NT-4	599235.1	4494572
		NT-5	599251.8	4494594
		NT-6	599251.6	4494605
		NT-7	599251	4494650
		NT-8	599267.8	4494661
		NT-9	599268.3	4494628
		NT-10	599293.3	4494650
		NT-11	599310.3	4494650
		NT-12	599309.8	4494684
		NT-13	599250.3	4494705
		NT-14	599249.9	4494738
Big Egg Control	Nekton	NC-15	599166.4	4494649
		NC-16	599149.1	4494682
		NC-17	599149.3	4494659
		NC-18	599140.9	4494659
		NC-19	599124.1	4494648
		NC-20	599099	4494625
		NC-21	598999.4	4494480
		NC-22	599024.6	4494491
		NC-23	599066.7	4494514
		NC-24	599109.3	4494492
		NC-25	599143.1	4494493
		NC-26	599168.5	4494493
Horseshoe Cove	Nekton	P1	584894	4478041
		P2	584842	4478039
		P3	584824	4478033
		P4	584795	4478041

Marsh	Sampling Variable	Station	UTM X (east)	UTM Y (north)
Horseshoe Cove (continued)	Nekton (continued)	P5	584744	4478051
		P6	584746	4478092
		P19	584700	4478127
		P7	584767	4478116
		P8	584788	4478130
		P9	584807	4478132
		P10	584811	4478169
		P11	584807	4478191
		P13	584770	4478211
		P14	584729	4478115
		P15	584706	4478213
		P16	584671	4478184
		P17	584646	4478171
		P18	584663	4478129
Horseshoe Cove	Vegetation	1-00	584884	4478098
		1-50	584827	4478089
		1-100	584780	4478076
		1-150*	584735	4478063
		1-200	584685	4478045
		1-250	584630	4478036
		2-00	584838	4478131
		2-50*	584791	4478122
		2-100	584738	4478109
		2-150	584690	4478096
		2-200	584641	4478087
		3-00	584814	4478181
		3-50	584758	4478176
		3-100	584710	4478175
		3-150*	584663	4478165
		3-200	584613	4478156
		4-00	584783	4478216
		4-50	584735	4478206
		4-100	584687	4478197
		4-150	584639	4478186
		4-200	584591	4478174

Table 6. Coordinates for locations sampled at SAIR in 2004, UTM, Zone 19, NAD 83, meters. ** Indicates UTM coordinates of stations were estimated from GIS maps because original GPS coordinates did not match up with adjacent stations.*

Sampling Variable	Station	UTM X (east)	UTM Y (north)
Nekton	1	334999	4703811
	2	334992	4703782
	3	334977	4703769
	4	334951	4703710
	5	334936	4703699
	6	334955	4703667
	7	334948	4703615
	8	334963	4703575
	9	334979	4703568
	10	335000	4703549
	11	335046	4703508
	12	335075	4703541
	13	335107	4703557
	14	335149	4703519
	15	335159	4703496
	16	335192	4703443
Vegetation	1A-00	335013	4703817
	1B-00	334986	4703818
	1B-10*	334976	4703818
	2B-00	334990	4703790
	2B-10*	334983	4703790
	2B-20	334977	4703789
	2B-30	334968	4703789
	2B-40	334959	4703789
	3B-00	334977	4703781
	3B-10	334968	4703781
	3B-20	334961	4703781
	3B-30*	334952	4703781
	4B-00	334964	4703761
	4B-10	334957	4703761
	4B-20*	334949	4703761
	4B-30	334941	4703761
	5A-00	334955	4703672
	5B-00*	334939	4703671
	5A-10*	334962	4703672

Sampling Variable	Station	UTM X (east)	UTM Y (north)
	5A-20	334970	4703673
	5A-30	334978	4703670
	5A-40	334990	4703671
	6A-00	334953	4703613
	6A-10	334964	4703615
	6A-20	334977	4703619
	6A-30	334984	4703617
	6A-40	334991	4703621
	7A-00	335009	4703554
	7A-10*	335017	4703559
	7A-20	335025	4703565

Table 7. Coordinates for locations sampled at SAHI in 2004, UTM, Zone 18, NAD 83, meters. * *Indicates UTM coordinates of stations were estimated from GIS maps because original GPS coordinates did not match up with adjacent stations.*

Sampling Variable	Station	UTM X (east)	UTM Y (north)
Nekton	1	627222	4527063
	2	627176	4527080
	3	627177	4527059
	4	627153	4527049
	5	627131	4527081
	6	627135	4527113
	7	627127	4527148
	8	627109	4527178
	9	627092	4527211
	11	627104	4527082
Vegetation	1-00	627212	4527117
	1-20	627196	4527116
	1-40	627181	4527114
	1-60	627164	4527104
	2-00	627209	4527152
	2-20	627180	4527148
	2-40	627176	4527141
	2-60*	627156	4527135
	3-00	627195	4527168
	3-20	627180	4527172
	3-40	627158	4527161
	4-00*	627194	4527217
	4-20*	627183	4527215
	4-40*	627171	4527212
	4-60*	627160	4527210
	4-80*	627147	4527207
	5-00	627096	4527065
	6-00	627086	4527092
	6-20	627111	4527086
	6-40	627130	4527102
	6-60	627149	4527104
	7-00	627092	4527113
	7-20	627116	4527124
	7-40	627138	4527130
	7-60	627148	4527144
	8-00	627098	4527149

Sampling Variable	Station	UTM X (east)	UTM Y (north)
	8-20	627111	4527150
	8-40	627126	4527151
	9-00	627084	4527183
	9-20*	627123	4527202
	9-40*	627136	4527203